### **DRAFT**

# Water Quality Criteria Report for Prometryn

Phase III: Application of the pesticide water quality criteria methodology



Prepared for the Central Valley Regional Water Quality Control Board

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June 2016

### **Disclaimer**

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### List of acronyms and abbreviations

AF Assessment factor

APHA American Public Health Association

ASTM American Society for Testing and Materials

BAF Bioaccumulation Factor

BC Black carbon

BCF Bioconcentration Factor
BMF Biomagnification Factor
CAS Chemical Abstract Service

CDFG California Department of Fish and Game

CSIRO Commonwealth Scientific and Industrial Research Organization, Australia

CVRWQCB Central Valley Regional Water Quality Control Board

DPR Department of Pesticide Regulation

 $EC_x$  Concentration that affects x% of exposed organisms

FDA Food and Drug Administration

FT Flow-through test

GMAV Genus Mean Acute Value

 $IC_r$  Inhibition concentration; concentration causing x% inhibition

ICE Interspecies Correlation Estimation

IUPAC International Union of Pure and Applied Chemistry

K Interaction Coefficient K<sub>H</sub> Henry's law constant

 $K_{ow}$  Octanol-Water partition coefficient  $K_p$  or  $K_d$  Solid-Water partition coefficient

 $LC_x$  Concentration lethal to x% of exposed organisms

 $LD_x$  Dose lethal to x% of exposed organisms

LL Less relevant, Less reliable study
LOEC Lowest-Observed Effect Concentration

LOEL Lowest-Observed Effect Level LR Less relevant, Reliable study

MATC Maximum Acceptable Toxicant Concentration

N Not relevant or Not reliable study

n/a Not applicable

NOAEL No-Observed Adverse Effect Level NOEC No-Observed Effect Concentration

NR Not reported

OECD Organization for Economic Co-operation and Development

OSAR Quantitative Structure Activity Relationship

pK<sub>a</sub> Acid dissociation constant RL Relevant, Less reliable study RR Relevant and Reliable study

S Static test

SMAV Species Mean Acute Value

SR Static renewal test

SSD Species Sensitivity Distribution
TES Threatened and Endangered Species

US United States

USEPA United States Environmental Protection Agency

#### 1 Introduction

A methodology for deriving freshwater water quality criteria for the protection of aquatic life was developed by the University of California - Davis (TenBrook et al. 2009a). The need for a methodology was identified by the California Central Valley Regional Water Quality Control Board (CVRWQCB 2006) and findings from a review of existing methodologies (TenBrook & Tjeerdema 2006, TenBrook et al. 2009b). The UC-Davis methodology is currently being used to derive aquatic life criteria for several pesticides of particular concern in the Sacramento River and San Joaquin River watersheds. The methodology report (TenBrook et al. 2009a) contains an introduction (Chapter 1); the rationale of the selection of specific methods (Chapter 2); detailed procedure for criteria derivation (Chapter 3); and a criteria report for a specific pesticide (Chapter 4). This criteria report for prometryn describes, section by section, the procedures used to derive criteria according to the UC-Davis methodology. Also included are references to specific sections of the methodology procedure detailed in Chapter 3 of the report so that the reader can refer to the report for further details (TenBrook et al. 2009a).

### 2 Basic information

Chemical: Prometryn or prometryne (Fig. 1)

CAS: N,N'-bis(1-methylethyl)-6-(methylthio)-1,3,5-triazine-2,4-diamine

CAS Number: 7287-19-6 USEPA PC Code: 080805 CA DPR Chem Code: 502

IUPAC: 6-methylsulfanyl-2-N,4-N-di(propan-2-yl)-1,3,5-triazine-2,4-diamine

Chemical Formula: C<sub>10</sub>H<sub>19</sub>N<sub>5</sub>S

Figure 1 Structure of prometryn

(source: American Chemical Society, 2015)

Trade names: Caparol, Gesagard, Prometrex, Primatol Q and Mercasin

## 3 Physical-chemical data

Molecular Weight

241.356 (http://webbook.nist.gov/cgi/inchi/InChI%3D1S/C10H19N5S/c1-

6(2)11-8-13-9(12-7(3)4)15-10(14-8)16-5/h6-7H%2C1-5H3%2C(H2%2C11%2C12%2C13%2C14%2C15)

**Density** 

1.15 g/mL (PPDB 2008)

Water Solubility

33 mg/L at 25°C (Worthing & Hance 1990)

33 mg/L at 25°C (PPDB 2015) 26.55 mg/L at 25°C (USEPA 2015) 33 mg/L at 20°C (USEPA 2015)

Geometric mean: 31.25 mg/L

Melting Point

132.03°C (USEPA 2015) 119°C (USEPA 2015) 119°C (PPDB 2015)

Geometric mean: 123.19 °C

Vapor Pressure

0.13 mPa at 25°C (PPDB 2015) 2.47 mPa at 25°C (USEPA 2015) **Geometric mean:** 0.567 mPa 25 °C

Henry's constant (K<sub>H</sub>)

2.24 x 10 <sup>-2</sup> Pa m<sup>3</sup> mol<sup>-1</sup> (USEPA 2015) 1.21 x 10 <sup>-3</sup> Pa m<sup>3</sup> mol<sup>-1</sup> (USEPA 2015) 1.20 x 10 <sup>-3</sup> Pa m<sup>3</sup> mol<sup>-1</sup> (PPDB 2015) **Geometric mean:** 3.19 x 10 <sup>-3</sup> Pa m<sup>3</sup> mol<sup>-1</sup>

Organic Carbon Sorption Partition Coefficients (log K<sub>oc</sub>)

All values from USEPA 2015

2.8172.656

Geometric mean: 2.735

Log Kow

\*Values referenced from the BioByte Bio-Loom program (2015)

3.73 (USEPA 2015) 3.51 (USEPA 2015)

- 3.34 (PPDB 2015)
- 3.10 (Tomlin 1997\*)
- 3.34 (Finizio et al. \*)
- 2.99 (Liu et al. 1995\*)
- 3.25 (Finizio *et al.* 1997\*)
- 3.03 (Schaeffer et al. 1970\*)
- 2.99 (Donovan and Pescatore 2002\*)

Geometric mean: 3.25

#### **Bioconcentration Factor**

Table 1 Bioconcentration factors (BCF) for prometryn

NR: not reported; values are on a wet weight basis and are not lipid-normalized.

Species	BCF	Exposure	Reference
NR	53.51	NR	USEPA 2015
NR	85	NR	PPDB 2015
Lepomis	110, viscera	28 d	Forbis and Halls, 1988
macrochirus	54, fillet		

#### **Environmental Fate**

Table 2 Prometryn hydrolysis and photolysis and other degradation.

(NR: not reported).

	Half- life	Water	Temp (°C)	pН	Reference
	(h or d)				
Hydrolysis	Stable	Aqueous buffer	25	5, 7, 9	Lawrence
					1987a
	75 d	NR	NR	NR	Johnson
					1991
Aqueous	55 d*	*River water,	NR,	****8.17	Navarro et
Photolysis	66 d**	sunlight	greenhouse	<sup>†,††</sup> 8.05	al. 2004
-	68 d†	**River water,		<sup>‡,‡‡</sup> 6.66	
	216 d††	darkness			
	88 d‡	†Seawater,			
	263 d‡‡	sunlight			
		††Seawater,			
		darkness			
		‡Groundwater,			
		sunlight			
		‡‡Groundwater,			
		darkness			
	4.6 h*	*Distilled	30-35	*7.1	Evgenidou
	11.6 h**	**Lake water		**8.7	and Fytianos
	6.9 h†	†River water		†8.5	2002

	50.6 d*	*Lake water	22	* 8.45	Konstantinou
	51.7 d**	**River water		**7.62	et al. 2001
	54.6 d†	†Marine water		<b>†</b> 7.45	
	27.7 d††	††Groundwater		††7.22	
	32.4 d‡	‡Distilled water		<b>‡</b> 5.89	
	Stable	Aqueous buffer	25	7	Lawrence
					1987b
	Stable	Aqueous	NR	NR	Halama et al.
		solution			no date
Biodegradation	150 d*	NR	NR	NR	Johnson
*aerobic	360 d**				1991
**anaerobic					

# 4 Human and wildlife dietary values

There are no FDA action levels for prometryn in food (USFDA 2000) and there are no EPA pesticide tolerances set for any aquatic species (USEPA 2013a).

#### Wildlife LC<sub>50</sub> values (dietary) for animals with significant food sources in water

The US EPA Environmental Risk Assessment for the Reregistration of Prometryn (USEPA 1996) states that prometryn is practically nontoxic to birds for acute and subacute exposures. The reported acute oral  $LC_{50}$  for mallard exceeds 4,640 mg/kg (Beavers and Fink, 1977) and the subacute dietary  $LC_{50}$  is 42,766 mg/kg. The latter study was received under US EPA MRID 70686; however, as it was illegible it could not be rated using the methodology or used for criteria derivation. Fletcher 1984 reported a  $LC_{50}$  value in excess of 5,000 mg/kg. A pilot study by Fletcher and Pedersen 1988 concluded no adverse effects in any tested concentration; therefore the  $EC_{50}$  value exceeds the highest tested concentration of 1,000 mg/kg.

#### Wildlife dietary NOEC values for animals with significant food sources in water

A mallard feeding study resulted in no statistically significant reproductive effects at any concentrations tested, thus the NOEC (no observed effect concentration) is reported as > 500 mg/kg, which was the highest test concentration (Fletcher 1989). This chronic value is an order of magnitude lower than the acute values available (Fletcher 1984; Beavers and Fink 1977) and should therefore be adequately protective of wildlife with significant food sources in water.

### 5 Ecotoxicity data

Approximately 31 original studies on the effects of prometryn on aquatic life were identified and reviewed. In the review process, many parameters were rated for documentation and acceptability for each study, including, but not limited to: organism source and care, control description and response, chemical purity, concentrations tested,

water quality conditions, and statistical methods (see Tables 3.6, 3.7, 3.8 in TenBrook et al. 2009a). Single-species effects studies that were rated as relevant (R) or less relevant (L) according to the method (Table 3.6) were summarized in data summary sheets. Information in these summaries was used to evaluate each study for reliability, using the rating systems described in the methodology (Tables 3.7 and 3.8, section 3-2.2, TenBrook et al. 2009a), to give a reliability rating of reliable (R), less reliable (L), or not reliable (N).

Studies of the effects of prometryn on mallard ducks were rated for reliability using the terrestrial wildlife evaluation. Mallard studies rated as reliable (R) or less reliable (L) were used to consider bioaccumulation. Three studies for mallard duck rating R were located in the literature and are described in Section 4.

Copies of completed summaries for all aquatic studies are included in the Appendix of this report. All data rated as acceptable (RR) or supplemental (RL, LR, LL) for criteria derivation are summarized in Tables 3 - 10, found at the end of this report. Acceptable studies rated as RR are used for numeric criteria derivation, while supplemental studies rated as RL, LR or LL are used for evaluation of the criteria to check that they are protective of particularly sensitive species and threatened and endangered species. These considerations are reviewed in section 12 and 14 of this report, respectively. Studies that were rated not relevant (N) or not reliable (RN or LN) were not used for criteria derivation.

One mesocosm study was identified and reviewed. This study was rated R and is listed in Appendix A3. It was used as supporting data in Section 13 to evaluate the derived criteria to ensure that they are protective of ecosystems.

#### Evaluation of aquatic animal data

Using the data evaluation criteria (section 3-2.2, TenBrook et al. 2009a), four acute studies yielding four toxicity values from three taxa were judged reliable and relevant for acute criterion derivation (Tables 3-4). Seven acute toxicity animal values for seven taxa from seven studies were rated RL, LL, or LR and were used as supplemental information for evaluation of the derived acute criterion in the Sensitive Species section 12 (Table 5). Five chronic animal toxicity values from five studies were rated RR (Tables 7-8). Three chronic toxicity animal values from one study was rated RL, LL, or LR (Table 10).

#### Evaluation of aquatic plant data

Plant data were used to derive the chronic criterion instead of chronic animal data because prometryn is an herbicide and plants are the most sensitive taxa (section 3-4.3, TenBrook et al. 2009a). All plant studies were considered chronic because the typical endpoints of growth or reproduction are inherently chronic. Three studies yielding three plant toxicity values were rated RR for the chronic criterion derivation (Tables 6).

Plant studies are more difficult to interpret than animal data because a variety of endpoints may be used, but the significance of each one is less clear. In this methodology, only endpoints of growth or reproduction (measured by biomass) and tests lasting at least 24-h had the potential to be rated highly and used for criteria calculation, which is in accordance with standard methods (ASTM 2007a, 2007b; USEPA 1996). The plant studies were rated for quality using the data evaluation criteria described in the methodology (section 3-2.2, TenBrook et al. 2009a).

The endpoints for plant data are all categorized as growth inhibition and are relative to a control growth measurement. Depending on the plant it may have been measured by direct cell counts with a hemacytometer, cell counts with a spectrophotometer, cell counts with an electronic particle counter, chlorophyll concentration measured by absorbance, turbidity measured by absorbance, or number of fronds (*Lemna spp.*). In all cases, growth of exposed samples was compared statistically to controls.

### 6 Data reduction

Multiple toxicity values for prometryn for the same species were reduced down to one species mean acute value (SMAV) or one species mean chronic value (SMCV) according to procedures described in the methodology (section 3-2.4, TenBrook et al. 2009a). Acceptable acute and chronic data that were reduced, and the reasons for their exclusion, are shown in Tables 4 and 8, respectively. Reasons for reduction of data included: a test with a more sensitive exposure duration for the same species was available, flow-through tests are preferred over static tests, a test with a more sensitive life-stage of the same species was available, and tests with more sensitive endpoints were available. The final acute animal, chronic plant, and chronic animal data sets are shown in Tables 3, 6, and 7, respectively.

#### 7 Acute criterion calculation

An acute criterion was calculated with acute animal toxicity data only, because plant toxicity tests are always considered chronic (section 3-2.1.1.1, TenBrook et al. 2009a). Since acceptable acute toxicity values were not available from the five required taxa for a species sensitivity distribution, the acute criterion was calculated using the Assessment Factor (AF) procedure (section 3-3.3, TenBrook et al. 2009a). Prometryn is an organic pesticide, and the AFs given in the methodology (Table 3.13, TenBrook et al. 2009a) are the most specific AFs available for organic pesticides. The methodology points out that the AFs are limited in that they are based on organochlorine and one organophosphate pesticides, which are neurotoxic insecticides, while prometryn is an herbicide that inhibits photosynthesis. However, prometryn does exhibit toxicity to animals with an unclear mechanism and is an organic pesticide, thus, it is reasonable to use the AF procedure for prometryn.

The AFs given in the methodology will be used for prometryn with the understanding that AFs based on measured pesticide toxicity data are likely more accurate than choosing an arbitrary AF. The methodology points out that AFs are recognized as a conservative approach for dealing with uncertainty in assessing risks posed by chemicals (section 2-3.2, TenBrook et al. 2009a). Using an AF to calculate a criterion always involves a high degree of uncertainty and there is potential or under- or over-protection, which is strongly dependent on the representation of sensitive species in the available data set. The methodology instructs that the derived criterion should be compared to all available ecotoxicity data to ensure that it will be protective of all species (section 3-6.0, TenBrook et al. 2009a).

There are two available taxa in the acceptable (RR) data set shown in the in Table 3: planktonic crustacean (*Daphnia magna*) and Salmonidae (*Oncorhynchus mykiss*). Missing from the taxa requirements for use of a species sensitivity distribution (SSD) are a warm water fish, a benthic crustacean, and an insect. The AF method calculates the criterion by dividing the lowest SMAV from the acceptable (RR) data set by an AF, which is determined by the number of taxa available in the data set (section 3-3.3, TenBrook et al. 2009a). The lowest SMAV was the 96-h *Oncorhynchus mykiss* LC<sub>50</sub> value of 5,460 µg/L. This value was divided by an AF of 12 because there are acceptable data from two taxa (Table 17, Fojut et al. 2014). The acute value calculated using the AF represents an estimate of the median 5<sup>th</sup> percentile value of the SSD, which is the recommended acute value. The recommended acute value is divided by a factor of 2 to calculate the acute criterion (section 3-3.3, TenBrook et al. 2009a). Because the toxicity data used to calculate the criterion only reported three significant figures, the criterion is rounded to three significant figures (section 3-3.2.6, TenBrook et al. 2009a).

```
Acute value = lowest value in data set \div assessment factor = 5,460 mg/L \div 12 = 455 \mug/L 
 Acute criterion = acute value \div 2 = 455 \mug/L \div 2 = 227.5 \mu/L
```

Acute criterion =  $228 \mu g/L$ 

### 8 Chronic criterion calculation

Prometryn is an herbicide and the chronic data in Tables 6 and 7 demonstrate that plants are the most sensitive taxa; therefore, the procedure for derivation of the chronic criterion of an herbicide was followed (section 3-4.3, TenBrook et al. 2009a). Acceptable chronic toxicity values were not available for five different species of vascular plants or alga, so a distribution could not be fit to the available chronic toxicity data (part 1, section 3-4.3, TenBrook et al. 2009a). The methodology instructs that in the absence of acceptable data to fit a distribution, the chronic criterion is equal to the lowest NOEC from an important alga or vascular aquatic plant species that has measured concentrations

and a biologically relevant endpoint (part 2, section 3-4.3, TenBrook et al. 2009a). Acceptable toxicity data for the aquatic plant *Navicula pelliculosa* (Hughes 1992b) is shown in Table 6, and the NOEC value reported for this species serves as the chronic criterion.

Chronic criterion =  $0.562 \mu g/L$ 

## 9 Water Quality Effects

### 9.1 *Bioavailability*

Few studies were found regarding the bioavailability of prometryn. Only one study was found that pertained to the bioavailability to organisms in the water column. Hermosin et al. (1982) found that prometryn bioavailability was affected by NH<sub>3</sub> gas treatment of montmorillonite clay-herbicide complexes that were prepared under acidic conditions (pH 3.5). Prometryn that desorbed from the clay surfaces was biologically active to the green alga *Chlamydomonas* and its bioavailability increased when the clay-pesticide complex had been exposed to NH<sub>3</sub> gas. Ammonia gas treatment caused the clay-adsorbed prometryn to be deprotonated and displaced from the interlamellar space to external surfaces of the clay particles. Bioavailability appears to be related to protonation and physical accessibility on the clay surface. This could be pertinent as ammonia gas is used as a fertilizer in agricultural soils and could be used in conjunction with prometryn pesticide application.

No other information about bioavailability of prometryn in the water column that differentiates when prometryn is sorbed to solids, sorbed to dissolved solids, or freely dissolved was found. Until there is more information that discusses the bioavailability of these three phases, compliance must be based on the total concentration of prometryn in water (section 3-5.1, TenBrook et al. 2009a).

#### 9.2 Mixtures

Prometryn can occur in the environment with other herbicides of similar or different modes of action. Prometryn is an s-triazine pesticide that acts as a photosystem II (PSII) inhibitor. Other widely used herbicides, such as the phenylurea class, are also PSII inhibitors, but have different binding sites than the triazine herbicides. The concentration addition model and the non-additive interaction model are the only predictive mixture models recommended by the methodology (section 3-5.2, TenBrook et al. 2009a), so other models found in the literature will not be considered for compliance.

Several studies have confirmed that toxicity of a mixture of herbicides that are PSII-inhibitors can be predicted by the concentration addition method (Fuast et al. 2000 and 2001, Drost et al. 2003, Wilkinson et al. 2015). Faust et al. (2000) studied a mixture of 18 triazines with identical mechanisms of action with a unicellular green alga *Scenedesmus vacuolatus* and found that the combined toxicity could be predicted by

concentration addition. Faust et al. (2001) again used a mixture of 18 different s-traizine herbicides with unicellular green alga *Scenedesmus vacuolatus* to show that the toxic effects of the mixture exceeded that of the most active ingredient alone. Even nonsignificant effect concentrations of the herbicides contributed to mixture toxicity. Concentration addition predictions were accurate for all effect levels and concentration ratios of herbicides. Drost et al. (2003) reported that concentration addition prediction was valid for a mixture of four s-triazines with *Lemna minor*. Near complete recovery of growth occurred within three days when the plants were moved to pesticide-free growth medium. Concentration addition was also valid in seawater as reported by Wilkinson et al. (2015). In this study, a mixture of ten photosystem II herbicides of similar mechanism of action was tested on the seagrass *Halophila ovali*.

Trimble and Lydy (2006) studied the effect of prometryn on the organophosphate insecticide chlorpyrifos on the amphipod crustacean *Hyalella azteca*. Prometryn did not cause a significant effect on chlorpyrifos toxicity. A synergistic ratio of 1.10 was calculated in a binary mixture with chlorpyrifos.

In summary, when prometryn is detected with other s-triazine PSII-inhibitor herbicides the toxicity should be predicted by the concentration addition model. There are no multi-species coefficients of interaction reported in the literature, so the non-additive interaction model cannot be used to assess water quality criteria compliance when other types of contaminants are present. No studies on aquatic organisms were identified in the literature that could provide a quantitative means to consider mixtures of prometryn with other classes of pesticides.

### 9.3 Temperature, pH, and other water quality effects

Temperature, pH, and other water quality effects on the toxicity of prometryn were examined to determine if any effects are described well enough in the literature to incorporate into criteria compliance (section 3-5.3, TenBrook et al. 2009a). There were no studies available that examined the effects of temperature or pH on toxicity in the aqueous environment. As prometryn is a moderately weak base, pH is not expected to have a significant effect on the chemical structure in the range of conditions found in natural freshwater environments.

### 10 Comparison of ecotoxicity data to derived criteria

# 10.1 *Sensitive species*

The derived criteria were compared to toxicity values for the most sensitive species in both the acceptable (RR) and supplemental (RL, LR, LL) data sets to ensure that these species will be adequately protected (section 3-6.1, TenBrook et al. 2009a).

The lowest acute value in the data sets rated RR, RL, LR, or LL (Tables 3, 4, and 5) is 1,700 µg/L for mysid shrimp (*Mysidopsis bahia*), which is rated LR because it is a

saltwater species. The lowest freshwater acute value an LC50 of 2,500  $\mu$ g/L for *Oncorhynchus mykiss* (Beliles 1965). This study rated RL because water quality parameters were not reported and prometryn concentrations were not reported as either measured or nominal. The derived acute criterion (228  $\mu$ g/L) is also based on this species and is lower than the Beliles 1965 value and should protective of *O. mykiss*. The derived acute criterion of 228  $\mu$ g/L is lower than all other toxicity values in the data set, thus it is expected that sensitive animal species will be protected if the acute criterion concentration is attained.

The derived chronic criterion (0.562  $\mu$ g/L) is the lowest of all chronic data that was highly rated (Table 5) and is equal to the NOEC for growth inhibition of an aquatic plant. The next lowest acceptable value for another species is the NOEC of 8.1  $\mu$ g/L for microalga *Raphidocelis subcapitata* (Hughes 1987). Prometryn is an herbicide so it is expected that plants will be more sensitive than animals, therefore the chronic criterion should be adequately protective of both plant and animal species.

### 10.2 Ecosystem and other studies

The derived criteria are compared to acceptable laboratory, field, or semi-field multispecies studies (rated R or L) to determine if the criteria will be protective of ecosystems (section 3-6.2, TenBrook et al. 2009a). One mesocosm, microcosm or ecosystem (field and laboratory) study was identified. The laboratory microcosm study tested an alga (*Cryptomonas* sp.) and an algivorous ciliate (*Urotricha furcata*) alone and in combination and rated L (Liebig et al. 2008). Nominal exposure concentrations for the dual species microcosm and the single species tests were greater than both the acute and chronic criteria (ranging from 8.75-15,000  $\mu$ g/L). The NOEC related to area under the growth curve was 6.9  $\mu$ g/L for alga alone and 2,200  $\mu$ g/L for the ciliate alone. Both species had a NOEC of 15.2  $\mu$ g/L in the microcosm combination. The authors speculate that the much lower microcosm NOEC for the ciliate was an indirect effect of reduced alga for consumption rather than direct toxicity by prometryn.

Although limited to this single study, there is evidence that prometryn in aquatic ecosystems may have detrimental effects on the bottom of the food chain, which may indirectly impact species up the food chain via changes in water quality or decreased food supply. The derived chronic criterion of  $0.562~\mu g/L$  is protective of the alga or the ciliate based on the individual and combined NOECs available.

### 10.3 Threatened and endangered species

The derived criteria are compared to measured toxicity values for threatened and endangered species (TES), as well as to predicted toxicity values for TES, to ensure that they will be protective of these species (section 3-6.3, TenBrook et al. 2009a). Current lists of state and federally listed threatened and endangered plant and animal species in California were obtained from the California Department of Fish and Game website (CDFG 2015). One listed animal species is represented in the dataset. Five Evolutionarily Significant Units of *Oncorhynchus mykiss* are listed as federally threatened or endangered throughout California. The acute data set include one 96-hr LC<sub>50</sub> value for *O*.

mykiss of 5,460  $\mu$ g/L (Hamaker 1985b). A supplemental study that rated L primarily due to a lack of water quality parameter description reported a LC<sub>50</sub> of 2,500  $\mu$ g/L for *O. mykiss* (Beliles 1965). These data indicate that the acute criterion of 228  $\mu$ g/L would be protective of this species.

The USEPA interspecies correlation estimation (ICE v. 3.1; USEPA 2010) software was used to estimate toxicity values for the listed animals or plants represented in the acute data set by members of the same family or genus. Table 10 summarizes the results of the ICE analyses. The estimated toxicity values in Table 10 range from 6,630.85  $\mu$ g/L for Chinook salmon, 9,057.90  $\mu$ g/L Coho salmon, and 5278.47  $\mu$ g/L for cutthroat salmon. Prometryn toxicity values were out of range of the values used to develop the model for apache trout.

No plant studies used in the criteria derivation were of state or federal endangered, threatened or rare species. Plants are particularly sensitive to prometryn because it is an herbicide, but there are no aquatic plants listed as state or federal endangered, threatened or rare species so they could not be considered in this section.

Based on the available data and estimated values for animals, there is no evidence that the value referenced in place of a calculated acute and or the calculated chronic criteria will be underprotective of threatened and endangered species.

#### 11 Harmonization with other environmental media

#### 11.1 Bioaccumulation

Bioaccumulation was assessed to ensure that the derived criteria will not lead to unacceptable levels of prometryn in food items (section 3-7.1, TenBrook et al. 2009a). Prometryn has a log  $K_{ow}$  of 3.25 (BioByte 2015), a  $K_d$  of 0.9-45 depending on soil type (Baskaran and Kennedy 1999; Gawlick et al. 1999; Saxena 1987), and a molecular weight of 241.36, which indicates some degree of bioaccumulative potential. There are no FDA action levels for prometryn in food (USFDA 2000), and there are no EPA pesticide tolerances set for any aquatic species (USEPA 2013a). Bioconcentration of prometryn has been measured in an unknown species (Table 1).

To check that these criteria are protective of terrestrial wildlife that may consume aquatic organisms, a bioaccumulation factor (BAF) was used to estimate the water concentration that would roughly equate to a reported toxicity value for such terrestrial wildlife (LC<sub>50, oral predator</sub>). These calculations are further described in section 3-7.1 of the methodology (TenBrook et al. 2009a). The BAF of a given chemical is the product of the BCF and a biomagnification factor (BMF), such that BAF=BCF\*BMF. No BMF value was found for prometryn. Chronic dietary toxicity values are preferred for this calculation. The BAF and BCF values available were either from an estimation modeling program (USEPA 2015), the value origin was not reported (PPDB 2015), or from a chronic exposure study in a freshwater fish (Forbis and Halls 1988). The lowest dietary

value for mallard was > 500 mg/kg (Fletcher 1989). A value of 500 mg/kg with the BCF 72 L/kg (USEPA 2015, PPDB 2015, and Forbis and Halls 1998) were used as an example estimation of bioaccumulation in the environment. No BMF value was available in the literature so it was estimated two ways according to the methodology (a value of 1 both when as approximated from log  $K_{ow}$  and as approximated from BCF as in section 3-7.1 and Table 3.15 in TenBrook *et al.* 2009a).

$$NOEC_{water} = \frac{NOEC_{oral-predator}}{BCF_{food\_item} \cdot BMF_{food\_item}}$$

Mallard: 
$$NOEC_{water} = \frac{500^{mg}/kg}{72 \frac{L}{kg} * 1} = 6.94^{mg}/L = 6940^{\mu g}/L$$

In this example, the calculated chronic criterion (0.562  $\mu$ g/L) is more than three orders of magnitude below the estimated NOEC<sub>water</sub> value for wildlife and is not expected to cause adverse effects due to bioaccumulation.

#### 11.2 Harmonization with air and sediment criteria

This section addresses how the maximum allowable concentration of prometryn might impact life in other environmental compartments through partitioning (section 3-7.2, TenBrook et al. 2009a). The 2013 EPA Environmental Fate and Ecological Risk Assessment Problem Formulation in Support of the Registration Review of Prometryn indicates that chronic toxicity data of prometryn to sediment-dwelling organisms are required (USEPA 2013b). The only available sediment value for prometryn is estimated based on partitioning from water using empirical log  $K_{\rm oc}$  values. These range from 2.656  $\mu g/L$  to 2.817  $\mu g/L$  (USEPA 2015). Pesticides having a high log  $K_{\rm OC}$  sorb to the soil and are not transported into the water column. The value for prometryn is mid-range when compared to other pesticides (Delle Site 2001). Sorption reduces the bioavailability of prometryn to aquatic organisms but could adversely affect benthic organisms. There are no other federal or state sediment or air quality standards for prometryn (CARB 2008; CDWR 1995), nor is prometryn mentioned in the NOAA sediment quality guidelines (NOAA 1999). For biota, the limited data on bioconcentration or biomagnification of prometryn is addressed in section 15.

# 12 Prometryn criteria summary

# 12.1 Limitations, assumptions, and uncertainties

The assumptions, limitations and uncertainties involved in criteria generation are available to inform environmental managers of the accuracy and confidence in criteria (section 3-8.0, TenBrook et al. 2009a). Chapter 2 of the methodology (TenBrook et al.

2009a) discusses these points for each section as different procedures were chosen, such as the list of assumptions associated with using an SSD (section 2-3.1.5.1), and reviews them in section 2-7.0. This section summarizes any data limitations that affected the procedure used to determine the final prometryn criteria.

Overall, there was a lack a highly rated aquatic plant and animal toxicity data for prometryn. Both the acute and chronic data sets lacked the full complement of five required taxa to fit a distribution for criteria derivation. The acute data set was missing values for a warm water fish, a benthic crustacean, and an insect. The AF procedure was used to calculate the acute criterion. The chronic data set contained only three out of five different species of vascular plants or alga. The lowest NOEC from an important alga or vascular aquatic plant species was identified as the chronic criterion.

The most important limitation is the lack of acceptable plant data because prometryn is an herbicide. Plant and algal data is difficult to interpret and do not use consistent endpoints. The assumptions that went into evaluation of plant studies are described in section 5. The chronic data set only contained three plant values, precluding the use of a SSD, although all of the studies reported a NOEC, LOEC, and MATC, which are the appropriate toxicity values for chronic tests. The methodology requires that MATC values are used to derive chronic criterion by the SSD procedure, unless studies are available with EC<sub>x</sub> values that show what level of x is appropriate to represent a noeffect level (section 3-2.1.1.2, TenBrook et al. 2009a). The chronic criterion was derived with the absolute minimum amount of data according to the methodology (part 2, section 3-4.3, TenBrook et al. 2009a) as the lowest NOEC from an important alga or vascular aquatic plant, and uncertainty in the chronic criterion cannot be quantified because it is based on only one toxicity value.

Chronic animal taxa requirements were not met, as there were only three values available. However, chronic animal data is not used for chronic criterion derivation of an herbicide, or when plants are the most sensitive taxa to a particular pesticide (3-4.3, TenBrook et al. 2009a). Although prometryn is an herbicide, some animals do show sensitivity to it.

Other limitations include the lack of sediment studies to assess partitioning of prometryn from other environmental niches than the water column. Only one study was available, although the 2013 EPA Environmental Fate and Ecological Risk Assessment Problem Formulation in Support of the Registration Review of Prometryn called for studies testing the chronic toxicity of prometryn to sediment-dwelling organisms (USEPA 2013b). One bioavailability study was available for organisms in the water column. There is evidence that prometryn availability is affected through interaction with clay minerals in the presence of ammonia gas and results in toxicity to at least one alga species. Further studies are needed to determine the extent of this mineral-herbicide interaction. Additional mallard duck studies are needed to determine definitive toxicity values. The available studies that are highly rated reported estimates equal to the highest tested exposure concentrations. This information is not useful for criteria derivation.

### 12.2 Comparison to national standard methods

This section is provided as a comparison between the UC-Davis methodology for criteria calculation (TenBrook et al. 2009a) and the current USEPA (1985) national standard. The following example prometryn criteria were generated using the USEPA (1985) methodology with the data set generated in this prometryn criteria report.

The USEPA acute methods have three additional taxa requirements beyond the five required by the SSD procedure of the UC-Davis methodology (section 3-3.1, TenBrook et al. 2009a). They are:

- 1. A third family in the phylum Chordata (e.g., fish, amphibian);
- 2. A family in a phylum other than Arthropoda or Chordata (e.g., Rotifera, Annelida, Mollusca);
- 3. A family in any order of insect or any phylum not already represented.

Only one of the three additional requirements could be met with *P. leniusculus* in the Astacidae family. Missing from the USEPA (1985) methodology requirements are a warm water fish, a benthic crustacean, an insect, a third family in the phylum Chordata, and a family in a phylum other than Arthropoda or Chordata. Because of this lack of data, no acute criterion could be calculated according to the USEPA (1985) methodology.

According to the USEPA (1985) methodology, the chronic criterion is equal to the lowest of the Final Chronic Value, the Final Plant Value, and the Final Residue Value.

To calculate the Final Chronic Value, animal data is used and the same taxa requirements must be met as in the calculation of the acute criterion (section III B USEPA 1985). One of the eight taxa requirements is available in the RR chronic animal data set with *Cyprinus carpio L*. (Table 7). The missing taxa are as follows:

- 1. A benthic crustacean
- 2. An insect (aquatic exposure)
- 3. A third family in the phylum Chordata (e.g., fish, amphibian)
- 4. A family in a phylum other than Arthropoda or Chordata (e.g., Rotifera, Annelida, Mollusca)
- 5. A family in any order of insect or any phylum not already represented

The California Department of Fish and Game has derived criteria using the USEPA (1985) SSD method with fewer than the eight required families, using professional judgment to determine that species in the missing categories were relatively insensitive and their addition would not lower the criteria (Menconi & Beckman 1996; Siepmann & Jones 1998). In this case, there are too many missing taxa values to derive a Final Chronic Value in this way.

The Final Plant Value is calculated as the lowest result from a 96-hr test conducted with an important plant species in which the concentrations of test material were measured and the endpoint was biologically important. None of the plant toxicity

values in the RR data set (Table 6) are for a 96-hr test; they are longer ranging from five to 14 days. The lowest NOEC reported is 0.562 µg/L for *Navicula pelliculosa* (Hughes 1992b), which also serves as the derived chronic criterion. This test has an exposure duration that is four days longer than the specified duration.

Final Plant Value = lowest result from a plant test =  $0.562 \mu g/L$ 

The Final Residue Value is calculated by dividing the maximum permissible tissue concentration by an appropriate bioconcentration or bioaccumulation factor. A maximum allowable tissue concentration is either (a) a FDA action level for fish oil or for the edible portion of fish or shellfish, or (b) a maximum acceptable dietary intake based on observations on survival, growth, or reproduction in a chronic wildlife feeding study or long-term wildlife field study. There are no FDA action levels for prometryn in food (USFDA 2000) and there are no EPA pesticide tolerances set for any aquatic species (USEPA 2013a). A single dietary NOEC of 500 mg/kg (Fletcher 1989) was the lowest wildlife dietary toxicity value available. A BCF of 67 for an unknown species (Table 1) is used to calculate the Final Residue Value.

Final Residue Value = maximum acceptable dietary intake  $\div$  BCF = 500 mg/kg  $\div$  67 L/kg = 7.46 mg/L = 7,460  $\mu$ g/L

The Final Plant Value is lower than the Final Residue Value. A Final Chronic Value cannot be calculated. Therefore the chronic criterion by the USEPA (1985) methodology for prometryn would be  $0.562~\mu g/L$ . The example chronic criterion is equivalent to the one recommended by the UC-Davis methodology.

#### 12.3 Final criteria statement

The final criteria statement is:

Aquatic life in the Sacramento River and San Joaquin River basins should not be affected unacceptably if the four-day average concentration of prometryn does not exceed 0.562  $\mu$ g/L more than once every three years on the average and if the one-hour average concentration does not exceed 228  $\mu$ g/L more than once every three years on the average.

Although the criteria were derived to be protective of aquatic life in the Sacramento and San Joaquin Rivers, these criteria would be appropriate for any freshwater ecosystem in North America, unless species more sensitive than are represented by the species examined in the development of these criteria are likely to occur in those ecosystems.

The acute criterion is based only on acute animal data and was derived to protect animals from acute pulses of prometryn. Details of the acute criterion calculation are described in section 7 and the acute data are shown in Tables 3 - 5. An assessment factor was used instead of a distribution to calculate the acute criterion because there were not sufficient data from the five required taxa for use of a SSD.

Details of the chronic criterion calculation are described in section 8 and chronic plant data are shown in Table 6. The chronic criterion was derived to only be protective of plants, but will also likely be protective of animals, which are less sensitive to prometryn. The lowest NOEC of a highly rated plant study was used as the criterion because there were insufficient data for use of a SSD for criterion calculation. The chronic criterion was calculated with the absolute minimum amount of data, and uncertainty cannot be quantified. Plant toxicity data is essential when considering prometryn usage and regulations because plants and algae are the most sensitive taxa, however, plant data are difficult to interpret. The chronic criterion was derived using the best data available, and firm evidence that could support lowering criteria was not found. The criteria should be updated whenever new relevant and reliable data is available.

There are no established water quality criteria for prometryn with which to compare the criteria derived in this report. The US EPA has several aquatic life benchmarks established for prometryn, shown in Table 12, to which the derived criteria in this report can be compared with caution (USEPA 2014). According to the USEPA (2014), aquatic life benchmarks are not calculated following the same methodology used to calculate water quality criteria. Water quality criteria can be used to set water quality standards under the Clean Water Act, but aquatic life benchmarks may not be used for this purpose (USEPA 2014). The acute criterion of 228  $\mu g/L$  is well below both the acute fish benchmark of 1,455  $\mu g/L$  and the acute invertebrate benchmark of 4,850  $\mu g/L$  (by factors of 5 and 17 times, respectively). The derived chronic criterion of 0.562  $\mu g/L$  is well below the chronic benchmarks for fish and invertebrates of 620  $\mu g/L$  and 1000  $\mu g/L$ , respectively. The chronic criterion is approximately a factor of 2 below the acute nonvascular plant benchmark of 1.04  $\mu g/L$ . Because the chronic criterion was derived using only plant data, it is most comparable to the acute nonvascular plant benchmark.

The final acute criterion was derived using the AF procedure (section 7) and the acute data used in for the calculation are shown in Table 3. Due to a lack of acceptable data to fit a distribution, the chronic criterion is equal to the lowest NOEC from an important alga or vascular aquatic plant species (section 8). Chronic data rated RR are shown in Table 6. It is recommended that the whole water prometryn concentration is measured for water quality criteria compliance until additional bioavailability studies are available (section 9).

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**Data Tables** 

Table 3 Final acute toxicity data set for prometryn.

All studies were rated RR and were conducted at standard temperature. S: static; SR: static renewal; FT: flow-through.

Species	Common Identifier	Family	Test type	Meas/ Nom	Chemical grade	Duration	Temp (°C)	Endpoint	Age/ size	LC/EC <sub>50</sub> (μg/L) (95% CI)	Reference
Daphnia magna	Waterflea	Daphniidae	S	Meas	97.00%	48-h	20	Mortality	< 24-h	12,660 (7,569- 24,033)	Hamaker 1985a
Daphnia magna	Waterflea	Daphniidae	S	Nom	98.90%	48-h	17	Mortality	< 20-h	18,900 (16,000- 22,200)	Vilkas 1977
Daphnia magna										15,468	GEOMEAN
Oncorhynchus mykiss	Rainbow trout	Salmonidae	S	Meas	97.00%	96-h	12	Mortality	30-d post hatch	5,460 (5,183- 5,771)	Hamaker 1985b
Pacifastacus leniusculus	Signal crayfish	Astacidae	SR	Meas	99.30%	96-h	19	Mortality	5-8th stage	12,100	Velisek 2013

Table 4 Supplemental acute data rated RL, LR, LL.

Reason for exclusion given below. S: static; SR: static renewal; FT: flow-through. NR: not reported. 95% CI: 95% confidence interval. Exclusion reasons are listed at the end of the table.

	Common		Test	Meas/	Chemical		Temp			LC/EC <sub>50</sub> (µg/L) (95%		Rating/
Species	Identifier	Family	type	Nom	grade	Duration	(°C)	Endpoint	Age/ size	CI)	Reference	Reason
Mysidopsis bahia	Mysid shrimp	Mysidae	S	Meas	98.10%	96-h	25	Mortality	< 24-h	1700 (1400- 2000)	Surprenant 1988a	2
Carrassius auratus	Goldfish	Cyprinidae	SR	NR	99.00%	96-h	17.5	Mortality	1.1 g, 3.5 cm	3500 (530- 6600)	Beliles 1965	6
Cyprinodon variegatus	Sheepshead minnow	Cyprinodo ntidae	S	Meas	98.10%	96-h	22	Mortality	0.42 g, 29 mm	5100 (4000- 7000)	Surprenant 1988b	2
Daphnia magna	Waterflea	Daphniida e	S	NR	>96.00%	48-h	21	Immobilizati on	< 24-h	9700	Marchini 1988	5
Lepomis macrochirus	Bluegill sunfish	Centrarchi dae	SR	NR	99.00%	96-h	20.5	Mortality	0.8g, 3.4 cm	10000 (6200- 14000)	Beliles 1965	6
Mercenaria mercenaria	Quahog clam	Veneridae	S	Meas	98.10%	48-h	22	Normal larvae count	Embryo/ larvae	21000 (120- 51000)	Surprenant 1988c	2
Oncorhynchus mykiss	Rainbow trout	Salmonida e	SR	NR	99.00%	96-h	14.5	Mortality	0.9 g, 3.9 cm	2500 (1600- 4000)	Beliles 1965	6

#### **Exclusion Reasons**

- 1. Not a standard method
- 2. Saltwater
- 3. Low chemical purity or purity not reported
- 4. Toxicity value not calculable
- 5. Control response not reported
- 6. Low reliability score

Table 5 Final chronic plant toxicity data set for prometryn.

All studies were rated RR. S: static; SR: static renewal; FT: flow-through. NR: not reported, n/a: not applicable.

Species	Common identifier, Family	Test type	Meas/ Nom	Chemical grade	Duration	Temp (°C)	Endpoint	Age/ size	NOEC (μg/L)	LOEC (µg/L)	MATC (μg/L)	EC <sub>50</sub> (µg/L)	Reference
_							Growth inhibition					12.2	Hughes &
Lemna gibba	Duckweed	S	Meas	98.40%	14-d	25	(frond count)	7- 11-d	3.99	8.42	5.80	(10.6- 14.1)	Alexander 1992a
Navicula pelliculosa	Diatom	S	Meas	98.40%	5-d	24	Growth inhibition (cell count)	7-d	0.562	0.962	0.735	1.40 (1.12- 1.75)	Hughes & Alexander 1992b
Raphidoceli s subcapitata	Microalga	S	Meas	98.10%	7-d	24	Growth inhibition (cell count)	7-d	8.1	16	11	23	Hughes 1987
Anabena flos-aquae	Cyanobacterium	S	Meas	98.40%	5-d	24	Growth inhibition (cell count)	Alga l cells	20.2	35.2	26.7	40.5 (33.0- 49.7)	Hughes 1992

Table 6 Final chronic animal toxicity data set for prometryn.

All studies were rated RR. S: static; SR: static renewal; FT: flow-through. NR: not reported

Species	Common identifier	Test type	Chemical grade	Duration	Endpoint	Age/size	NOEC (μg/L)	LOEC (µg/L)	MATC (μg/L)	Reference
Cyprinus carpio L.	Common carp	SR	99.30%	35-d	Mortality	Fertilized eggs	850	1100	967	Velisek 2015
Daphnia magna	Waterflea	FT	98.10%	21-d	Growth, percent survival, reproduction	< 24-h	1000	2000	1400	Surprenant 1988d
Pimephales promelas	Fathead minnow	FT	98.40%	32-d	Mortality, hatching success, growth	< 24-h	620	1200	860	Graves 1995

Table 7 Acceptable reduced chronic data rated RR.

Reason for exclusion given below. S: static; SR: static renewal; FT: flow-through. NR: not reported

Species	Common identifier	Test type	Meas/ Nom	Chemical grade	Duration	Temp (°C)	Endpoint	Age/ size	NOEC (μg/L)	LOEC (µg/L)	MATC (μg/L)	Reference	Reason for exclusion
							Reproduction						
							(% embryos						
							producing						
							live fry at 32-						
							d); Growth						
Pimephales	Fathead						(Length and					Humaker	
promelas	minnow	FT	Meas	97.00%	32-d	20	weight)	< 48-h	802	1390	1056	1985c	A

A. Less sensitive life-stage

Table 8 Supplemental chronic plant data rated RL, LR, or LL.

S: static; SR: static renewal; FT: flow-through. NR: not reported, n/a: not applicable; 95% CI: 95% confidence interval; SE: standard error.

Species	Common identifier	Test type	Meas/ Nom	Chemical grade	Duration	Temp (°C)	Endpoint	Age/size	NOEC (µg/L)	LOEC (µg/L)	EC <sub>50</sub> (μg/L) (95% CI)	Reference	Rating/ Reason for exclusion
CLL II							Growth	A11					
Chlorella vulgaris	Green alga	S	Nom	77.13%	96-h	25	inhibition (cell count)	Algal cells	NR	NR	53.6*	Ma 2002	2, 3
vuigaris	Oreen arga	<u> </u>	NOIII	77.1370	90-11	23	Growth	Cells	INIX	IVIX	7.63	N1a 2002	2, 3
Skeleonema	Marine						inhibition	Algal			(6.86-	Hughes	
costatum	diatom	S	Meas	98.40%	5-d	20	(cell count)	cells	2.22	4.54	8.49)	1993	1
Costation	didioni		TVICUS	70.1070		20	Growth	cons	2.22	1.5 1	31.5	1773	
Cryptomonas							inhibition	Algal			(29.5-		
sp.	Alga	S	Meas	99.20%	7-d	20	(cell count)	cells	23.2	34.8	34.1)	Liebig 2008	4
·	_						Growth	Algal				_	
Cryptomonas							inhibition	cells		Not			
sp.	Alga	S	Meas	99.20%	14-d	20	(cell count)	cens	34.8	calculable	NR	Liebig 2008	4
							Growth						
							inhibition	Algal			22.0		
Cryptomonas							(area under	cells			22.9 (18.0-		
Cryptomonas	Alga	S	Meas	99.20%	7-d	20	growth curve)		6.9	10.3	32.1)	Liebig 2008	4
sp.	Aiga	S	wicas	99.2070	/-u	20	Growth		0.9	10.5	32.1)	Licoig 2006	4
							inhibition						
							(area under	Algal					
Cryptomonas							growth	cells					
sp.	Alga	S	Meas	99.20%	14-d	20	curve)		15.5	23.2	NR	Liebig 2008	4
							Growth	Algal			39.3		
Cryptomonas							inhibition	cells			(37.1-		
sp.	Alga	S	Meas	99.20%	7-d	20	(growth rate)	cens	23.2	34.8	42.4)	Liebig 2008	4

#### **Exclusion Reasons**

<sup>1.</sup> Saltwater

<sup>2.</sup> Low chemical purity or purity not reported

- 3. Low reliability score4. Control not described and/or response not reported

Table 9 Supplemental chronic animal data rated RL, LR, or LL.

S: static; SR: static renewal; FT: flow-through. NR: not reported.

Species	Common identifier	Test type	Meas/ Nom	Chemical grade	Duration	Temp (°C)	Endpoint	Age/size	NOEC (μg/L)	LOEC (µg/L)	MATC (μg/L) (95% CI)	Reference	Rating/ Reason for exclusion
Americamysis bahia	Saltwater mysid	FT	Meas	97.80%	28-d	25	Mortality	< 24-h	450	840	615	Claude 2013	2
Americamysis bahia	Saltwater mysid	FT	Meas	97.80%	28-d	25	Reproduction	< 24-h	110	220	156	Claude 2013	2
Americamysis bahia	Saltwater mysid	FT	Meas	97.80%	28-d	25	Growth (Length and weight)	< 24-h	450	840	615	Claude 2013	2
Procambarus fallax f. virginalis	Marbled crayfish	S	Meas	99.30%	53-d	12	Mortality	Early lifestages	0.1	0.51	0.23	Velisek 2014	1, 3
Procambarus fallax f. virginalis	Marbled crayfish	S	Meas	99.30%	53-d	12	Mortality	Early lifestages			EC50: 40	Velisek 2014	1, 3

#### **Exclusion Reasons**

- 1. Not a standard method
- 2. Saltwater
- 3. Control response not reported

Table 10 Threatened, Endangered, or Rare Species Predicted values by ICE.

Surr	ogate		Predicted
	$LC_{50}$		$LC_{50}$ (95% confidence interval)
<b>Species</b>	$(\mu g/L)$	Species	$(\mu g/L)$
Rainbow	5460	_	
trout		Chinook salmon (O. tshawytscha)	6630.85 (3543.92-12406.6)
		Coho salmon (O. kisutch)	9057.90 (6801.88-12062.19)
		Cutthroat trout (O. clarkii)	5278.47 (3907.55-7130.36)
		Apache trout (O. gilae)	Surrogate $LC_{50}$ outside range for prediction model

Table 11 US EPA Aquatic Life Benchmarks.						
All units are μg/I	All units are μg/L. (USEPA 2014)					
Acute Fish	Chronic Fish	Acute	Chronic	Acute		
		Invertebrates	Invertebrates	nonvascular		
plants						
1455	620	4850	1000	1.04		

# **Appendix A - Aqueous Toxicity Data Summaries**

# **Appendix A1 - Aqueous Toxicity Studies Rated RR**

#### Anabena flos-aquae

Study: Hughes JS, Alexander MM. (1992c) The toxicity of prometryn technical to *Anabena flosaquae*. Malcom Pirnie, Inc., White Plains, New York. Laboratory project ID B267-577-1. Ciba-Geigy Corporation, Greensboro, NC. USEPA MRID 42520902.

RelevanceReliabilityScore: 100Score: 96Rating: RRating: R

Relevance points taken off for: none.

	<b>Hughes &amp; Alexander 1992c</b>	A. flos-aquae
Parameter	Value	Comment
Test method cited	Laboratory protocol B267- 577-1 which satisfies EPA's Pesticide Assessment Guidelines	
Phylum/subphylum	Cyanobacteria	
Order	Nostocales	
Family	Nostocaceae	
Genus	Anabena	
Species	Flos-aquae (Lyng.) Breb.	
Family native to North America?	Yes	
Age/size at start of test/growth phase	Algal cells	
Source of organisms	Laboratory cultures	
Have organisms been exposed to contaminants?	No	
Animals acclimated and disease-free?	Yes	
Animals randomized?	Not reported	Given organism size and presence in growth medium, it is assumed that aliquots are inherently randomly
Test vessels randomized?	Yes	
Test duration	5 d	
Data for multiple times?	3, 4, 5 d	
Effect 1	Cell count	
Control response 1	3 d: 24,000 4 d: 123,667 5 d: 340,000	
Temperature	24 ± 2 °C	
Test type	Static	

	<b>Hughes &amp; Alexander 1992c</b>	A. flos-aquae
Parameter	Value	Comment
Photoperiod/light intensity	Continuous; 2153 lux	
Dilution water	Growth medium	Synthetic algal assay procedure nutrient medium made with Type I water
рН	7.5	
Feeding	Growth medium	
Purity of test substance	98.4 %	
Concentrations measured?	Yes	
Measured is what % of nominal?	70.4-129 %	
Toxicity values calculated based on nominal or measured concentrations?	Measured	
Chemical method documented?	GC	
Concentration of carrier (if any) in test solutions	Dimethylformamide, 0.4 mL/L	
Concentration 1 Nom; Meas (µg/L)	2.3; 2.53	3,000 cells/mL/rep, 3 reps
Concentration 2 Nom; Meas (µg/L)	4.60; 5.32	3,000 cells/mL/rep, 3 reps
Concentration 3 Nom; Meas (µg/L)	9.20; 7.46	3,000 cells/mL/rep, 3 reps
Concentration 4 Nom; Meas (µg/L)	18.4;20.2	3,000 cells/mL/rep, 3 reps
Concentration 5 Nom; Meas (µg/L)	36.8; 35.2	3,000 cells/mL/rep, 3 reps
Concentration 6 Nom; Meas (µg/L)	73.6; 59.3	3,000 cells/mL/rep, 3 reps
Control	Negative: 0; 0	3,000 cells/mL/rep,
	Solvent: 0; 0	3 reps
EC <sub>25</sub> (95% CI) (μg/L)	25.6 (17.9-36.4)	Method:
EC <sub>50</sub> (95% CI) (μg/L)	40.5 (33.0-49.7)	Method: Non-linear regression
NOEC	20.2	Method: Dunnett's test p: 0.05 MSD: not reported
LOEC	35.2	Method: Dunnett's test
MATC	26.7	
% control at NOEC	3 d: 70 %	3 d: 11,333 (tmt) /
	4 d: 50 % 5 d: 47 %	16167 (mean controls) = 70 %
		4 d: 42,333 (tmt) / 84,834 (mean

	<b>Hughes &amp; Alexander 1992c</b>	A. flos-aquae
Parameter	Value	Comment
		controls) = 50 %
		5 d: 109,333 (tmt) /
		234,167 (mean
		controls) = 47 %
% control at LOEC	5 d: 31 %	5 d: 71,667 (tmt) /
		234,166.5 (mean
		controls) = 31 %

Prometryn solubility (S) = 31,250 ug/L  $\mu$ g/L, 2S = 62,500  $\mu$ g/L.

Reliability points were not taken off for water quality parameters (hardness, alkalinity, conductivity) because there is no guidance for these parameters in the test guidelines for algal/plant studies, the growth medium used requires distilled water, and the medium is presumably appropriate for the test species because a specific culture media was used.

Reliability points taken off for:

<u>Documentation:</u> Minimum significant difference (2). Total: 100-2 = 98

<u>Acceptability:</u> Temperature variation (3), Minimum significant difference (1), % control at NOEC (1), % control at LOEC (1). Total: 100- 6=94

Reliability score: mean(98, 94)=96

# Cyprinus carprio L.

Study: Velisek J, Stara A, Koutnik D, Machova J. (2015) Effects of prometryne on early life stages of common carp (*Cyprinus carpio* L.) Pesticide Biochemistry and Physiology. 118: 58-63.

RelevanceReliabilityScore: 100Score: 83Rating: RRating: R

Relevance points taken off for: none

	Velisek et al. 2015	C. carpio
Parameter	Value	Comment
Test method cited	Modified test No. 210: Fish,	
	Early Life Stage Toxicity	
	Test OECD.	
Phylum/subphylum	Chordata	
Class	Actinopterygii	
Order	Cypriniformes	
Family	Cyprinidae	
Genus	Cyprinus	
Species	Carpio	
Family native to North America?	Introduced	
Age/size at start of test/growth	Fertilized eggs	
phase		
Source of organisms	Faculty of Fisheries and	
	Protection of Waters,	
	Vodnany, Czech Republic	
Have organisms been exposed to	No	
contaminants?		
Animals acclimated and disease-	Yes	
free?		
Animals randomized?	No	
Test vessels randomized?	No	
Test duration	35 d	
Data for multiple times?	Some data for 14 d	
Effect 1	Survival	
Control response 1	86%	
Effect 2	Mass	
Control response 2	106.73 mg	
Effect 3	Length	
Control response 3	19 mm (Fig 3)	
Effect 4	Growth rate	
Control response 4	12.93	
Effect 5	Hatching	
Control response 5	Not reported	

ek et al. 2015  C. carpio  Comment
yo viability
ported
0.95 °C
renewal Dilution water changed daily
ported
ed tap water
1
ported
ported
ported
6
6 d, freshly hatched
shrimp Artemia salina
i ad libitum
6
ported
r · · · ·
S/MS
not reported 3 reps, 100/rep
t reported
not reported
not reported
reported
Survival: 2,314 Method: probit
ated/extrapolated)
Survival: 850 Method: probit
Survival: 1,100 Method: probit
,
Survival: 80 Method: one-way
Hatching: 80 ANOVA
Embryo viability: 80 p: 0.01
nass: 1200 MSD: not reported
ength: 1200
Survival: 1200 Same as above
Statching: 1200
Embryo viability: 1200
nass: 4000
ength: 4000
urvival: 310
Statching: 310

	Velisek et al. 2015	C. carpio
Parameter	Value	Comment
	35 d Embryo viability: 310	
	35 d mass: 2,191	
	35 d length: 2,191	
% control at NOEC	35 d Survival:	
	87%/86%=101% (Fig 1)	
	35 d Hatching: not	
	calculable	
	35 d Embryo viability: not	
	calculable	
	35 d mass: 85/106.73 mg=	
	80%	
	35 d	
	length:18.5/19mm=97%	
% control at LOEC	35 d Survival:	
	73%/86%=85% (Fig 1)	
	35 d Hatching: not	
	calculable	
	35 d Embryo viability: not	
	calculable	
	35 d mass: 38/106.73	
	mg=36%	
	35 d length:	
	13.5/19mm=71%	

Prometryn solubility (S) = 31,250 ug/L  $\mu$ g/L, 2S = 62,500  $\mu$ g/L. All exposure concentrations were acceptable.

#### Reliability points taken off for:

<u>Documentation:</u> Measured concentrations (3), Hardness (2), Alkalinity (2), Conductivity (2), Photoperiod (3), Minimum significant difference (2), % control at NOEC/LOEC (2). Total: 100 - 16 = 84

<u>Acceptability:</u> Organisms randomized (1), Hardness (2), Alkalinity (2), Conductivity (1), Photoperiod (2), Number of concentrations (3), Random design (2), Dilution factor (2), Hypothesis tests (3). Total: 100 - 18 = 82

Reliability score: mean (84, 82) = 83

#### Daphnia magna

Study: Hamaker TL. (1985a) *Daphnia magna* 48-hour static acute toxicity test with prometryn. Environmental Research and Technology, Inc., Fort Collins, CO study number D187. Submitted to Syngenta Crop Protection, LLC, Greensboro, NC. USEPA MRID 491390-03.

RelevanceReliabilityScore: 100Score: 89.5Rating: RRating: R

Relevance points taken off for: none

	Hamaker 1985a	D. magna
Parameter	Value	Comment
Test method cited	Standard Practice for	
	Conducting Acute Toxicity	
	Tests with Fishes,	
	Macroinvertebrates, and	
	Amphibians (ASTM 1980).	
Phylum/subphylum	Arthropoda/Crustacea	
Class	Branchiopoda	
Order	Cladocera	
Family	Daphniidae	
Genus	Daphnia	
Species	magna	
Family native to North America?	Yes	
Age/size at start of test/growth	< 24 h	
phase		
Source of organisms	ERT Bioassay Laboratory	
	stock cultures	
Have organisms been exposed to	No	
contaminants?		
Animals acclimated and disease-	Yes	
free?		
Animals randomized?	Yes	
Test vessels randomized?	Yes	
Test duration	48 h	
Data for multiple times?	Yes	24, 48 h
Effect 1	Mortality	
Control response 1	100% survival	
Temperature	20 ± 1 °C	
Test type	Static	
Photoperiod/light intensity	16 l: 8 d	
Dilution water	Reconstituted	
рН	8	
Hardness	100 mg/L CaCO <sub>3</sub>	

	Hamaker 1985a	D. magna
Parameter	Value	Comment
Alkalinity	$72 \pm 2 \text{ mg/L}$	
Conductivity	$339 \pm 12 \text{ umhos/cm}$	
Dissolved Oxygen	$7 \pm 3 \text{ mg/L}$	44-110 %
Feeding	Not reported	
Purity of test substance	97 %	
Concentrations measured?	Yes	
Measured is what % of nominal?	88-106	
Toxicity values calculated based on	Measured	
nominal or measured		
concentrations?		
Chemical method documented?	GC	
Concentration of carrier (if any) in	250 mL/L	Acetone
test solutions		
Concentration 1 Nom; Meas (µg/L)	5000; 5210	2 reps, 5/rep
Concentration 2 Nom; Meas (µg/L)	7000; 6940	2 reps, 5/rep
Concentration 3 Nom; Meas (µg/L)	10000; 8790	2 reps, 5/rep
Concentration 4 Nom; Meas (µg/L)	14000; 14820	2 reps, 5/rep
Concentration 5 Nom; Meas (µg/L)	20000; 19820	2 reps, 5/rep
Control	Negative: 0; 0	2 reps, 5/rep
	Solvent: 0; 0	
EC <sub>50</sub> (95% CI) (μg/L)	48 h:12,660 (7,569-24,033)	Method: probit

Notes: Acetone carrier concentration 500 times greater than allowed for acute tests.

Prometryn solubility (S) = 31,250 ug/L  $\mu$ g/L, 2S = 62,500  $\mu$ g/L. All exposure concentrations were acceptable.

#### Reliability points taken off for:

<u>Documentation:</u> Statistical significance (2), Significance level (2), Minimum significant difference (2), % control at NOEC/LOEC (2). Total: 100 - 8 = 92

<u>Acceptability:</u> Dissolved oxygen (6), Carrier solvent (4), Minimum significant difference (1), % control at NOEC (1), % control at LOEC (1). Total: 100 - 13 = 87

Reliability score: mean (92, 87) = 89.5

#### Daphnia magna

Study: Surprenant DC. (1988d) The chronic toxicity of prometryn technical to *Daphnia magna* under flow-through conditions. Springborn Life Sciences, Inc., Wareham, MA. Laboratory study number 88-1-2622. Ciba-Geigy Corporation, Greensboro, NC. USEPA MRID 405737-20.

RelevanceReliabilityScore: 100Score: 95.5Rating: RRating: R

Relevance points taken off for: none

	Surprenant 1988d	D. magna
Parameter	Value	Comment
Test method cited	Protocol for Conducting a	Springborn Life
	Flow-Through Life Cycle	Sciences protocol
	Toxicity Test with Daphnia	
	magna (# 081087/DM.LC)	
Phylum/subphylum	Arthropoda/Crustacea	
Class	Branchiopoda	
Order	Cladocera	
Family	Daphniidae	
Genus	Daphnia	
Species	magna	
Family native to North America?	Yes	
Age/size at start of test/growth	≤ 24 h	
phase		
Source of organisms	Laboratory culture	
Have organisms been exposed to	No	
contaminants?		
Animals acclimated and disease-	Yes	
free?		
Animals randomized?	Yes	
Test vessels randomized?	Not reported	
Test duration	21 d	
Data for multiple times?	1, 2, 4, 7, 14, 21 d	
Effect 1	Growth	
Control response 1	Length: 4.2 mm	
Effect 2	Percent survival	
Control response 2	7 d: 93	
	14 d: 96	
	21 d: 93	
Effect 3	Reproductive performance	Number offspring
Control response 3	14 d: 17	
	21 d: 42 ± 8.7	
Temperature	20 ± 1 °C	

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	Surprenant 1988d	D. magna
Parameter	Value	Comment
	21 d: 86 %	
		Reproduction:
	Length:	36 (tmt) / 42
	21d: 83 %	(control) = 86 %
		Length:
		3.5 (tmt) / 4.2
		(control) = 83 %
% control at LOEC	Survival:	LOEC 21 d (1000)
	21 d: 98 %	Survival:
		91 (tmt) / 93
	Reproduction:	(control) = 98 %
	21 d: 98 %	
		Reproduction:
	Length:	41 (tmt) / 42
	21d: 90 %	(control) = 98 %
		T41
		Length:
		3.8 (tmt) / 4.2
		(control) = 90 %

Prometryn solubility (S) = 31,250 ug/L  $\mu$ g/L, 2S = 62,500  $\mu$ g/L so all exposure concentrations were acceptable.

Reliability points taken off for:

<u>Documentation</u>: Statistical significance (2), Significance level (2), Minimum significant difference (2). Total: 100 - 6 = 94

Acceptability: Random design (2), Minimum significant difference (1). Total: 100 - 3 = 97

Reliability score: mean (94, 97) = 95.5

**Water Toxicity Data Summary** 

D. magna

Study: Vilkas AG. (1977) Acute toxicity of prometryn-FL-761355 to the water feal *Daphnia magna* straus. Union Carbide Environmental Services, Tarrytown, NY. Project number 11506-04-04. Prepared for Ciba-Geigy Corporation, Greensboro, NC. CADPR study ID 952592.

RelevanceReliabilityScore: 100Score: 78Rating: RRating: R

	Vilkas 1977	D. magna
Parameter	Value	Comment
Test method cited	Practices of The Committee	
	on Methods for Toxicity	
	Tests with Aquatic	
	Organisms, 1975	
Phylum/subphylum	Arthropoda/Crustacea	
Class	Branchiopoda	
Order	Cladocera	
Family	Daphniidae	
Genus	Daphnia	
Species	magna	
Family native to North America?	Yes	
Age/size at start of test/growth	First instar, < 20 h	
phase		
Source of organisms	National Water Quality	
	Laboratory, Duluth,	
	Minnesota	
Have organisms been exposed to	No	
contaminants?		
Animals acclimated and disease-	Yes	20 h
free?		
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	48 h	
Data for multiple times?	Yes	24, 48 h
Effect 1	Mortality	
Control response 1	0	
Temperature	17 ± 1 °C	
Test type	Static	
Photoperiod/light intensity	Not reported	
Dilution water	Small lake in Westchester	Filtered prior to use
	County, New York	
рН	7.21	
Hardness	50 mg/L CaCO <sub>3</sub>	
Alkalinity	26 mg/L CaCO <sub>3</sub>	
Conductivity	110 umhos/cm	
Dissolved Oxygen	8.1-9.7 mg/L	84-100 %
Feeding	Not fed	

	Vilkas 1977	D. magna
Parameter	Value	Comment
Purity of test substance	98.9 %	Not reported but found purity of prometryn-FL- 761355 on USEPA website
Concentrations measured?	Not reported	
Measured is what % of nominal?	Not reported	
Toxicity values calculated based on nominal or measured concentrations?	Nominal	
Chemical method documented?	No	
Concentration of carrier (if any) in test solutions	Not reported	
Concentration 1 Nom; Meas (µg/L)	10000	4 reps, 5/rep
Concentration 2 Nom; Meas (µg/L)	32000	4 reps, 5/rep
Concentration 3 Nom; Meas (µg/L)	100000, > 2S	4 reps, 5/rep
Control	Negative: 0 Solvent: 0	Solvent: acetone at concentration equal to amount in highest concentration
LC <sub>50</sub> (95% CI) (μg/L)	18900 (16000, 22200)	Method: Spearman- Karber Estimator
NOEC	< 10000	Method: p: MSD:
% control at NOEC	Not calculable	

Notes: Prometryn solubility (S) = 31,250 ug/L  $\mu$ g/L, 2S = 62,500  $\mu$ g/L. One exposure concentration exceeded 2S.

In adequate number of concentrations tested (3), UCDM requires  $\geq 5$ .

# Reliability points taken off for:

<u>Documentation</u>: Analytical method (4), Measured concentrations (3), Photoperiod (3), Statistical significance (2), Significance level (2), Minimum significant difference (2), % control at NOEC/LOEC (2). Total: 100 - 18 = 82

<u>Acceptability:</u> Measured concentrations within 20% nominal (4), Concentrations not > 2x solubility (4), Carrier solvent (4), Organisms randomized (1), Temperature (3), Photoperiod (2), Number of concentrations (3), Random design (2), Minimum significant difference (1), % control at NOEC (1), % control at LOEC (1). Total: 100 - 26 = 74

Reliability score: mean (82, 74) = 78

**Water Toxicity Data Summary** 

Lemna gibba

Study: Hughes JS, Alexander MM. (1992a) The toxicity of prometryn technical to *Lemna gibba G3*. Malcolm Pirnie, Inc., Tarrytown, NY. Laboratory study ID B267-577-4. Ciba-Geigy Corporation, Greensboro, NC. USEPA MRID 42520901.

RelevanceReliabilityScore: 100Score: 95Rating: RRating: R

Relevance points taken off for: None

	Hughes & Alexander 1992a	L. gibba
Parameter	Value	Comment
Test method cited	MPI Protocol No. B267-577-4	Based on ASTM protocol (1991), satisfies EPA Pesticide Assessment Guidelines (Holst and Ellwanger, 1982)
Order	Alismatales	
Family	Araceae	
Genus	Lemna	
Species	gibba	
Family native to North America?	Yes	
Age/size at start of test/growth phase	7-11 d	
Source of organisms	Laboratory stock cultures	
Have organisms been exposed to contaminants?	No	
Animals acclimated and disease-free?	Yes	
Animals randomized?	Not reported	Aliquots of cultures used so essentially randomized due to size of organisms.
Test vessels randomized?	Not reported	
Test duration	14 d	
Data for multiple times?	3, 5, 7, 10, 12, 14 d	
Effect 1	Frond count	
Control response 1	Negative: 33 Solvent: 38	
Temperature	25 ± 2 °C	
Test type	Static	
Photoperiod/light intensity	4198-5813 lumens/m <sup>3</sup>	
Dilution water	Growth medium	20X-AAP, 20 strength synthetic algal assay procedure nutrient medium

	<b>Hughes &amp; Alexander 1992a</b>	L. gibba
Parameter	Value	Comment
pН	8.52	Mean
Feeding	Growth medium	
Purity of test substance	98.4 %	
Concentrations measured?	Yes	
Measured is what % of nominal?	70-100 %	
Toxicity values calculated based on nominal or measured concentrations?	Measured	
Chemical method documented?	GC	
Concentration of carrier (if any) in test solutions	DMF 0.5 mL/L	
Concentration 1 Nom; Meas (µg/L)	1.25; 1.01	3 plants (12 fronds)/rep; 3 reps
Concentration 2 Nom; Meas (µg/L)	2.50; 1.76	3 plants (12 fronds)/rep; 3 reps
Concentration 3 Nom; Meas (µg/L)	5.00; 3.99	3 plants (12 fronds)/rep; 3 reps
Concentration 4 Nom; Meas (µg/L)	10.0; 8.42	3 plants (12 fronds)/rep; 3 reps
Concentration 5 Nom; Meas (µg/L)	20.0; 18.1	3 plants (12 fronds)/rep; 3 reps
Concentration 6 Nom; Meas (µg/L)	40.0; 40.2	3 plants (12 fronds)/rep; 3 reps
Control	Negative: 0; Solvent:	3 plants (12 fronds)/rep; 3 reps
EC <sub>25</sub> (95% CI) (μg/L)	14 d: 6.70 (5.40-8.31)	Method: weighted least squares non-linear regression
EC <sub>50</sub> (95% CI) (μg/L)	14 d: 12.2 (10.6-14.1)	Method: weighted least squares non-linear regression
NOEC	3.99	Method: ANOVA and Dunnett's test p: 0.05 MSD: Not reported
LOEC	8.42	
MATC	5.80	
% control at NOEC	14 d: 88 %	NOEC=3.99 14 d: 499 (tmt) / 567.5 (mean controls) = 88
% control at LOEC	14 d: 68 %	14 d: 384 (tmt) / 567.5 (mean controls) = 68

Prometryn solubility (S) = 31,250 ug/L  $\mu$ g/L, 2S = 62,500  $\mu$ g/L so all test exposure concentrations were acceptable.

Reliability points were not taken off for water quality parameters (hardness, alkalinity, conductivity) because there is no guidance for these parameters in the test guidelines for algal/plant studies, the growth medium used requires distilled water, and the medium is presumably appropriate for the test species because a specific culture media was used.

Reliability points taken off for:

Documentation: Minimum significant difference (2). Total: 100 - 2 = 98

<u>Acceptability:</u> Carrier solvent (4), Random design (2), Minimum significant difference (1), % control at LOEC (1). Total: 100 - 8 = 92

Reliability score: mean (98, 92) = 95

## Navicula pelliculosa

Study: Hughes JS, Alexander MM. (1992b) The toxicity of prometryn technical to *Navicula pelliculosa*. Malcom Pirnie, Inc., White Plains, New York. Laboratory project ID 0267-38-1100-1. Ciba-Geigy Corporation, Greensboro, NC. USEPA MRID 42620201.

RelevanceReliabilityScore: 100Score: 96.5Rating: RRating: R

	Hughes & Alexander 1992b	N. pelliculosa
Parameter	Value	Comment
Test method cited	MPI Protocol No. B267-577-2	Based on EPA protocol and satisfies EPA Pesticide Assessment Guidelines
Division	Heterokontophyta	
Class	Bacillariophyceae	
Order	Naviculales	
Family	Naviculaceae	
Genus	Navicula	
Species	pelliculosa	
Family native to North America?	Yes	
Age/size at start of test/growth phase	7 d	
Source of organisms	Laboratory stock cultures	
Have organisms been exposed to contaminants?	No	
Animals acclimated and disease-free?	Yes	
Animals randomized?	Not reported	Aliquots of culture removed from stock solutions during transfer to test vessel. Given size of organism, aliquots are assumed random.
Test vessels randomized?	Yes	Randomly repositioned
Test duration	5 d	-
Data for multiple times?	3, 4, 5 d	
Effect 1	Cell count/biomass	
Control response 1	5 d: 3.24 x 10 <sup>5</sup>	
Temperature	24 ± 2 °C	
Test type	Static	

	Hughes & Alexander 1992b	N. pelliculosa
Parameter	Value	Comment
Photoperiod/light intensity	4306 ± 464 lux, continuous	0 0222220
Dilution water	Growth medium	Synthetic algal assay procedure/Si nutrient medium, type 1 water
рН	$7.5 \pm 0.1$	<b>71</b>
Feeding	Growth medium	
Purity of test substance	98.4 %	
Concentrations measured?	Yes	
Measured is what % of nominal?	93.5 – 115 %	
Toxicity values calculated based on nominal or measured concentrations?	Measured	
Chemical method documented?	GC	
Concentration of carrier (if any) in test solutions	0.5 mL/L	
Concentration 1 Nom; Meas (µg/L)	0.25; 0.288	639 cells/rep, 4 reps
Concentration 2 Nom; Meas (µg/L)	0.50; 0.562	639 cells/rep, 4 reps
Concentration 3 Nom; Meas (µg/L)	1.00; 0.962	639 cells/rep, 4 reps
Concentration 4 Nom; Meas (µg/L)	2.00; 1.87	639 cells/rep, 4 reps
Concentration 5 Nom; Meas (µg/L)	4.00; 3.85	639 cells/rep, 4 reps
Concentration 6 Nom; Meas (µg/L)	8.00; 8.02	639 cells/rep, 4 reps
Control	Negative: 0; 0 Solvent: 0; 0	639 cells/rep, 4 reps 639 cells/rep, 4 reps
EC <sub>25</sub> (95% CI) (μg/L)	5 d: 0.890 (0.650, 1.22)	Method: weighted least squares nonlinear regression
EC <sub>50</sub> (95% CI) (μg/L)	5 d: 1.40 (1.12-1.75)	Method: weighted least squares nonlinear regression
NOEC (μg/L)	5 d: 0.562	Method: ANOVA and Dunnett's test p: 0.05 MSD: not reported
LOEC (µg/L)	5 d: 0.962	
MATC (geomean NOEC, LOEC) (μg/L)	5 d: 0.735	

	<b>Hughes &amp; Alexander 1992b</b>	N. pelliculosa
Parameter	Value	Comment
% control at NOEC	73.9% (based on solvent control)	1,182,565 (tmt) / 1,719,600 (mean controls) = 69%
% control at LOEC	71.8%	

Prometryn solubility (S) = 31,250 ug/L  $\mu$ g/L, 2S = 62,500  $\mu$ g/L so all test exposure concentrations were acceptable.

Reliability points were not taken off for water quality parameters (hardness, alkalinity, conductivity) because there is no guidance for these parameters in the test guidelines for algal/plant studies, the growth medium used requires distilled water, and the medium is presumably appropriate for the test species because a specific culture media was used. Points were not removed for dissolved oxygen since the test organism is an algae, which produces oxygen.

Reliability points taken off for:

Documentation: Minimum significant difference (2). Total: 100 - 2 = 98

<u>Acceptability:</u> Temperature variation (3), Minimum significant difference (1), % control at LOEC (1). Total: 100-5 = 95

Reliability score: mean (98, 95) = 96.5

# Oncorhynchus mykiss

Study: Hamaker TL. (1985b) Rainbow trout 96-hour static acute toxicity test with prometryn. Environmental Research and Technology, Inc., Fort Collins, CO study number TK0178833. Submitted to Ciba-Geigy Corporation, Greensboro, NC. USEPA MRID 490766-01.

RelevanceReliabilityScore: 100Score: 88Rating: RRating: R

	Hamaker 1985b	O. mykiss
Parameter	Value	Comment
Test method cited	Standard Practice for	
	Conducting Acute Toxicity	
	Tests with Fishes,	
	Macroinvertebrates, and	
	Amphibians, ASTM, 1980	
Phylum/subphylum	Chordata	
Class	Actinopterygii	
Order	Salmoniformes	
Family	Salmonidae	
Genus	Oncorhynchus	
Species	mykiss	
Family native to North America?	Yes	
Age/size at start of test/growth	Juvenile, 30 d post-hatch	
phase	1.5 g	
	3.0 cm	
Source of organisms	Cline Trout Farm, Boulder,	
	Colorado	
Have organisms been exposed to contaminants?	No	
Animals acclimated and disease-	Yes	
free?		
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	96 h	
Data for multiple times?	No	
Effect 1	Mortality	
Control response 1	100% survival	
Temperature	12 ± 1 °C	
Test type	Static	
Photoperiod/light intensity	16 l: 8 d	
Dilution water	Decholorinated, filtered	
	municipal water	
pH	$7.8 \pm 0.3$	
Hardness	28 mg/L CaCO <sub>3</sub>	

	Hamaker 1985b	O. mykiss
Parameter	Value	Comment
Alkalinity	Not reported	
Conductivity	Not reported	
Dissolved Oxygen	$7.55 \pm 1.5$	57-84 %
Feeding	Not reported	
Purity of test substance	97 %	
Concentrations measured?	Yes	
Measured is what % of nominal?	81-123	
Toxicity values calculated based on	Measured	
nominal or measured		
concentrations?		
Chemical method documented?	GC	Done by outside
		laboratory,
		Analytical
		Development
		Corporation,
		Monument,
		Colorado
Concentration of carrier (if any) in	Not reported	Acetone
test solutions		
Concentration 1 Nom; Meas (µg/L)	625; 680	2 reps, 10/rep
Concentration 2 Nom; Meas (µg/L)	1250; 1540	2 reps, 10/rep
Concentration 3 Nom; Meas (µg/L)	2500; 2550	2 reps, 10/rep
Concentration 4 Nom; Meas (µg/L)	5000; 4770	2 reps, 10/rep
Concentration 5 Nom; Meas (µg/L)	10000; 8160	2 reps, 10/rep
Control	Negative: 0; 0	2 reps, 10/rep
	Carrier: 0; 0	2 reps, 10/rep
LC <sub>50</sub> (95% CI) (µg/L)	5460 (5183-5771)	Method: Moving
_		average method

Prometryn solubility (S) = 31,250 ug/L  $\mu$ g/L, 2S = 62,500  $\mu$ g/L.

Reliability points taken off for:

<u>Documentation:</u> Alkalinity (2), Conductivity (2), Minimum significant difference (2), % control at NOEC/LOEC (2). Total: 100 - 8 = 92

<u>Acceptability:</u> Carrier solvent (4), Organisms randomized (1), Feeding (3), Hardness (2), Conductivity (1), Random design (2), Minimum significant difference (1), % control at NOEC (1), % control at LOEC (1). Total: 100 - 16 = 84

Reliability score: mean (92, 84) = 88

## Pacifastacus leniusculus

Study: Velisek, J. (2013) Acute toxicity of triazine pesticides to juvenile signal crayfish (*Pacifastacus leniusculus*). Neuroendrocrinology Letters. 34: 31-36.

RelevanceReliabilityScore: 100Score: 76.5Rating: RRating: R

	Velisek 2013	P. leniusculus
Parameter	Value	Comment
Test method cited	OECD No. 203	
Phylum/subphylum	Anthropoda/Custracea	
Class	Malacostraca	
Order	Decapoda	
Family	Astacidae	
Genus	Pacifastacus	
Species	leniusculus	
Family native to North America?	Yes	
Age/size at start of test/growth phase	5-8 <sup>th</sup> stage, 49.0-81.5 mg mass and 12.8-16.0 mm length)	
Source of organisms	Laboratory reared	
Have organisms been exposed to contaminants?	No	
Animals acclimated and disease-free?	72 h	
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	96 h	
Data for multiple times?	24, 48, 72, 96 h	
Effect 1	Mortality	
Control response 1	0%	
Temperature	$18.85 \pm 0.4  ^{\circ}\text{C}$	
Test type	Static renewal	Renewal every 48 h
Photoperiod/light intensity	Not reported	
Dilution water	Source not specified	
рН	7.50-7.82	
Hardness	Not reported	
Alkalinity	Not reported	
Conductivity	Not reported	
Dissolved Oxygen	95-100 %	
Feeding	Ad libitum to the 5 <sup>th</sup> -8 <sup>th</sup>	Chironomids and
	stage	pond zooplankton
Purity of test substance	99.3 %	
Concentrations measured?	Yes	

	Velisek 2013	P. leniusculus
Parameter	Value	Comment
Measured is what % of nominal?	≤5 %	
Toxicity values calculated based on nominal or measured concentrations?	Measured	
Chemical method documented?	HPLC, Katsumata et al. 2005	
Concentration of carrier (if any) in test solutions	<0.01 % DMSO	
Concentration 1 Nom; Meas (µg/L)	1000; not reported	3 reps, unknown/rep
Concentration 2 Nom; Meas (µg/L)	10,000; not reported	3 reps, unknown/rep
Concentration 3 Nom; Meas (µg/L)	40,000; not reported	3 reps, unknown/rep
Concentration 4 Nom; Meas (µg/L)	70,000; not reported; >2S	3 reps, unknown/rep
Concentration 5 Nom; Meas (µg/L)	100,000; not reported; >2S	3 reps, unknown/rep
Control	Dilution water; solvent	3 reps, unknown/rep
LC <sub>50</sub> (95% CI) (μg/L)	24 h: 76,800; > 2S 48 h: 17,000 72 h: 14,700 96 h: 12,100	Method: Linear regression w/probit

Notes: Confidence limits were not included in the study, although the toxicity values are reported "within 95% confidence limits."

Prometryn solubility (S) = 31,250 ug/L  $\mu$ g/L, 2S = 62,500  $\mu$ g/L. 24 h LC<sub>50</sub> exceeds 2S.

#### Reliability points taken off for:

<u>Documentation:</u> Measured concentrations (3), Dilution water (3), Dissolved oxygen (4), Conductivity (2), Photoperiod (3), Hypothesis tests (8). Total: 100 - 23 = 77

<u>Acceptability:</u> Concentrations not > 2x solubility (4), Organisms randomized (1), Adequate organisms per rep (2), Dilution water (2), Hardness (2), Alkalinity (2), Temperature (3), Conductivity (1), Photoperiod (2), Dilution factor (2), Random design (2), Dilution factor (2), Hypothesis tests (3). Total: 100 - 24 = 76

Reliability score: mean (77, 76) = 76.5

# Pimephales promelas

Study: Graves WC, Mank MA, Swigert JA. (1995) An early life-stage toxicity test with the fathead minnow (*Pimephales promelas*). Wildlife International, Ltd., Easton, Maryland. Laboratory study no. 108A-162. Ciba-Geigy Corporation, Greensboro, NC. USEPA MRID 43801702.

RelevanceReliabilityScore: 100Score: 93Rating: RRating: R

	Graves et al. 1995	P. promelas
Parameter	Value	Comment
Test method cited	Protocol based on Series 72 of Pesticide Assessment Guidelines, Subdivision E, Hazard Evaluation: Wildlife and Aquatic Organisms; ASTM Standard E1241-88, Standard Guide for Conducting Early Life-Stage Toxicity Tests with Fishes; Standard Evaluation Procedure, Fish Early Life-	Comment
DI 1 / 1 1 1	Stage Test	
Phylum/subphylum	Chordata	
Class	Actinopterygii	
Order	Cypriniformes	
Family	Cyprinidae	
Genus	Pimephales	
Species	promelas	
Family native to North America?	Yes	
Age/size at start of test/growth phase	< 24 h embryos	
Source of organisms	Chesapeake Cultures, Hayes, Virginia	
Have organisms been exposed to contaminants?	No	
Animals acclimated and disease-free?	Yes	
Animals randomized?	Not reported	
Test vessels randomized?	Yes	
Test duration	32 d	
Data for multiple times?	1, 2, 3, 4, 5, 6, 7, 14, 21, 28, 32 d	
Effect 1	Survival	

Parameter         Value         Comment           Control response 1         96 %         Mean controls           Effect 2         Hatching success           Control response 2         94.5 %         Mean controls           Effect 3         Wet weight         Control response 3         72.9 mg         Mean controls           Effect 4         Dry weight         Control response 4         16.5 mg         Mean controls           Effect 5         Length         Control response 5         19 mm         Mean controls           Temperature         25 ± 1 °C         Test type         Flow-through         Photoperiod/light intensity         I6l:8d, 336 lux         Dilution water         Publishing the state of the state o		Graves et al. 1995	P. promelas
Effect 2	Parameter		
Effect 2	Control response 1	96 %	Mean controls
Control response 2   94.5 %   Mean controls	•	Hatching success	
Method	Control response 2		Mean controls
Control response 3   72.9 mg	1	Wet weight	
Effect 4         Dry weight         Mean controls           Control response 4         16.5 mg         Mean controls           Effect 5         Length         Mean controls           Control response 5         19 mm         Mean controls           Temperature         25 ± 1 °C         Mean controls           Test type         Flow-through         Flow-through           Photoperiod/light intensity         16l:8d, 336 lux           Dilution water         Well water           pH         8           Hardness         135 mg/L CaCO₃         Mean day 32           Alkalinity         182 mg/L         Mean           Conductivity         338 umhos/cm         Mean           Dissolved Oxygen         6.4 mg/L         77 %           Feeding         Live brine shrimp nauplii (Artemia sp.) 3/d during first 7 d post hatch; 8-26 d post hatch; 8-26 d post hatch fed 2-3/d           Purity of test substance         98.4 %         Measured is what % of nominal?           Toxicity values calculated based on nominal or measured is what % of nominal?         Measured           Toxicity values calculated based on nominal or measured concentrations?         GC           Concentration I Nom; Meas (µg/L)         78; 81         40/rep, 2 reps           Concentration 1 Nom; Meas (µg/L)	Control response 3	Š	Mean controls
Control response 4		-	
Length   Control response 5   19 mm   Mean controls	Control response 4		Mean controls
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$		-	
Temperature			Mean controls
Test type	-		
Photoperiod/light intensity			
Dilution water   PH		$\mathcal{C}$	
pH         8           Hardness         135 mg/L CaCO₃         Mean day 32           Alkalinity         182 mg/L         Mean           Conductivity         338 umhos/cm         Mean           Dissolved Oxygen         6.4 mg/L         77 %           Feeding         Live brine shrimp nauplii (Artemia sp.) 3/d during first 7 d post hatch; 8-26 d post hatch fed 2-3/d         77 %           Purity of test substance         98.4 %         98.4 %         78 (Artemia sp.) 3/d during first 7 d post hatch; 8-26 d post hatch fed 2-3/d           Measured is what % of nominal?         Yes         Measured is what % of nominal?         Measured is what % of nominal?         Measured           Toxicity values calculated based on nominal or measured concentrations?         Measured         Measured           Chemical method documented?         GC         Acetone, 0.3 mL/L           Concentration 1 Nom; Meas (µg/L)         78; 81         40/rep, 2 reps           Concentration 2 Nom; Meas (µg/L)         160; 160         40/rep, 2 reps           Concentration 4 Nom; Meas (µg/L)         310; 310         40/rep, 2 reps           Concentration 5 Nom; Meas (µg/L)         1300; 1200         40/rep, 2 reps           Concentration 6 Nom; Meas (µg/L)         2500; 2400         40/rep, 2 reps           Control         Negative: 0;	Dilution water	,	
Hardness			
Alkalinity	1		Mean day 32
Conductivity   338 umhos/cm   Mean   77 %		2	•
Dissolved Oxygen  Feeding  Live brine shrimp nauplii (Artemia sp.) 3/d during first 7 d post hatch; 8-26 d post hatch fed 2-3/d  Purity of test substance  98.4 %  Concentrations measured?  Measured is what % of nominal?  Toxicity values calculated based on nominal or measured concentrations?  Chemical method documented?  Concentration of carrier (if any) in test solutions  Concentration 1 Nom; Meas (µg/L)  Concentration 2 Nom; Meas (µg/L)  Concentration 3 Nom; Meas (µg/L)  Concentration 4 Nom; Meas (µg/L)  Concentration 5 Nom; Meas (µg/L)  Concentration 6 Nom; Meas (µg/L)  Conc		<u> </u>	
Feeding  Live brine shrimp nauplii (Artemia sp.) 3/d during first 7 d post hatch; 8-26 d post hatch fed 2-3/d  Purity of test substance  98.4 %  Concentrations measured?  Measured is what % of nominal?  Toxicity values calculated based on nominal or measured concentrations?  Chemical method documented?  Concentration of carrier (if any) in test solutions  Concentration 1 Nom; Meas (µg/L)  Concentration 2 Nom; Meas (µg/L)  Concentration 3 Nom; Meas (µg/L)  Concentration 4 Nom; Meas (µg/L)  Concentration 5 Nom; Meas (µg/L)  Concentration 5 Nom; Meas (µg/L)  Concentration 6 Nom; M	·		
Cartemia sp.) 3/d during first 7 d post hatch; 8-26 d post hatch fed 2-3/d			77 70
first 7 d post hatch; 8-26 d post hatch fed 2-3/d  Purity of test substance  98.4 %  Concentrations measured?  Measured is what % of nominal?  Toxicity values calculated based on nominal or measured concentrations?  Chemical method documented?  Concentration of carrier (if any) in test solutions  Concentration 1 Nom; Meas (µg/L)  Concentration 2 Nom; Meas (µg/L)  Concentration 3 Nom; Meas (µg/L)  Concentration 4 Nom; Meas (µg/L)  Concentration 5 Nom; Meas (µg/L)  Concentration 6 Nom; Meas (µg/L)  Concentration	Teeding		
post hatch fed 2-3/d Purity of test substance  Oncentrations measured?  Measured is what % of nominal?  Toxicity values calculated based on nominal or measured concentrations?  Chemical method documented?  Concentration of carrier (if any) in test solutions  Concentration 1 Nom; Meas (μg/L)  Concentration 2 Nom; Meas (μg/L)  Concentration 3 Nom; Meas (μg/L)  Concentration 4 Nom; Meas (μg/L)  Concentration 5 Nom; Meas (μg/L)  Concentration 6 No			
Purity of test substance       98.4 %         Concentrations measured?       Yes         Measured is what % of nominal?       92-104 %         Toxicity values calculated based on nominal or measured concentrations?       Measured         Chemical method documented?       GC         Concentration of carrier (if any) in test solutions       Acetone, 0.3 mL/L         Concentration 1 Nom; Meas (μg/L)       78; 81       40/rep, 2 reps         Concentration 2 Nom; Meas (μg/L)       160; 160       40/rep, 2 reps         Concentration 3 Nom; Meas (μg/L)       310; 310       40/rep, 2 reps         Concentration 4 Nom; Meas (μg/L)       630; 620       40/rep, 2 reps         Concentration 5 Nom; Meas (μg/L)       1300; 1200       40/rep, 2 reps         Concentration 6 Nom; Meas (μg/L)       2500; 2400       40/rep, 2 reps         Control       Negative: 0;       40/rep, 2 reps         NOEC       620       Method: Batlett's test or Scheffe's multiple comparison test p: 0.05			
Concentrations measured?       Yes         Measured is what % of nominal?       92-104 %         Toxicity values calculated based on nominal or measured concentrations?       Measured         Chemical method documented?       GC         Concentration of carrier (if any) in test solutions       Acetone, 0.3 mL/L         Concentration 1 Nom; Meas (μg/L)       78; 81       40/rep, 2 reps         Concentration 2 Nom; Meas (μg/L)       160; 160       40/rep, 2 reps         Concentration 3 Nom; Meas (μg/L)       310; 310       40/rep, 2 reps         Concentration 4 Nom; Meas (μg/L)       630; 620       40/rep, 2 reps         Concentration 5 Nom; Meas (μg/L)       1300; 1200       40/rep, 2 reps         Concentration 6 Nom; Meas (μg/L)       2500; 2400       40/rep, 2 reps         Control       Negative: 0; Solvent: 0;       40/rep, 2 reps         NOEC       620       Method: Batlett's test or Scheffe's multiple comparison test p: 0.05	Purity of test substance		
Measured is what % of nominal?92-104 %Toxicity values calculated based on nominal or measured concentrations?MeasuredChemical method documented?GCConcentration of carrier (if any) in test solutionsAcetone, 0.3 mL/LConcentration 1 Nom; Meas (μg/L)78; 8140/rep, 2 repsConcentration 2 Nom; Meas (μg/L)160; 16040/rep, 2 repsConcentration 3 Nom; Meas (μg/L)310; 31040/rep, 2 repsConcentration 4 Nom; Meas (μg/L)630; 62040/rep, 2 repsConcentration 5 Nom; Meas (μg/L)1300; 120040/rep, 2 repsConcentration 6 Nom; Meas (μg/L)2500; 240040/rep, 2 repsControlNegative: 0; Solvent: 0;40/rep, 2 repsNOEC620Method: Batlett's test or Scheffe's multiple comparison test p: 0.05			
Toxicity values calculated based on nominal or measured concentrations?  Chemical method documented?  Concentration of carrier (if any) in test solutions  Concentration 1 Nom; Meas (μg/L) 78; 81 40/rep, 2 reps  Concentration 2 Nom; Meas (μg/L) 160; 160 40/rep, 2 reps  Concentration 3 Nom; Meas (μg/L) 310; 310 40/rep, 2 reps  Concentration 4 Nom; Meas (μg/L) 630; 620 40/rep, 2 reps  Concentration 5 Nom; Meas (μg/L) 1300; 1200 40/rep, 2 reps  Concentration 6 Nom; Meas (μg/L) 2500; 2400 40/rep, 2 reps  Control Negative: 0; 40/rep, 2 reps  NOEC 620 Method: Batlett's test or Scheffe's multiple comparison test p: 0.05			
nominal or measured concentrations?         GC           Chemical method documented?         GC           Concentration of carrier (if any) in test solutions         Acetone, 0.3 mL/L           Concentration 1 Nom; Meas (μg/L)         78; 81         40/rep, 2 reps           Concentration 2 Nom; Meas (μg/L)         160; 160         40/rep, 2 reps           Concentration 3 Nom; Meas (μg/L)         310; 310         40/rep, 2 reps           Concentration 4 Nom; Meas (μg/L)         630; 620         40/rep, 2 reps           Concentration 5 Nom; Meas (μg/L)         1300; 1200         40/rep, 2 reps           Concentration 6 Nom; Meas (μg/L)         2500; 2400         40/rep, 2 reps           Control         Negative: 0;         40/rep, 2 reps           NOEC         620         Method: Batlett's test or Scheffe's multiple comparison test p: 0.05			
Chemical method documented?GCConcentration of carrier (if any) in test solutionsAcetone, 0.3 mL/LConcentration 1 Nom; Meas (μg/L)78; 8140/rep, 2 repsConcentration 2 Nom; Meas (μg/L)160; 16040/rep, 2 repsConcentration 3 Nom; Meas (μg/L)310; 31040/rep, 2 repsConcentration 4 Nom; Meas (μg/L)630; 62040/rep, 2 repsConcentration 5 Nom; Meas (μg/L)1300; 120040/rep, 2 repsConcentration 6 Nom; Meas (μg/L)2500; 240040/rep, 2 repsControlNegative: 0; Solvent: 0;40/rep, 2 repsNOEC620Method: Batlett's test or Scheffe's multiple comparison test p: 0.05			
Chemical method documented?GCConcentration of carrier (if any) in test solutionsAcetone, 0.3 mL/LConcentration 1 Nom; Meas (μg/L)78; 8140/rep, 2 repsConcentration 2 Nom; Meas (μg/L)160; 16040/rep, 2 repsConcentration 3 Nom; Meas (μg/L)310; 31040/rep, 2 repsConcentration 4 Nom; Meas (μg/L)630; 62040/rep, 2 repsConcentration 5 Nom; Meas (μg/L)1300; 120040/rep, 2 repsConcentration 6 Nom; Meas (μg/L)2500; 240040/rep, 2 repsControlNegative: 0; Solvent: 0;40/rep, 2 repsNOEC620Method: Batlett's test or Scheffe's multiple comparison test p: 0.05	concentrations?		
Concentration of carrier (if any) in test solutionsAcetone, 0.3 mL/LConcentration 1 Nom; Meas (μg/L)78; 8140/rep, 2 repsConcentration 2 Nom; Meas (μg/L)160; 16040/rep, 2 repsConcentration 3 Nom; Meas (μg/L)310; 31040/rep, 2 repsConcentration 4 Nom; Meas (μg/L)630; 62040/rep, 2 repsConcentration 5 Nom; Meas (μg/L)1300; 120040/rep, 2 repsConcentration 6 Nom; Meas (μg/L)2500; 240040/rep, 2 repsControlNegative: 0; Solvent: 0;40/rep, 2 repsNOEC620Method: Batlett's test or Scheffe's multiple comparison test p: 0.05	Chemical method documented?	GC	
test solutions         Concentration 1 Nom; Meas (μg/L)         78; 81         40/rep, 2 reps           Concentration 2 Nom; Meas (μg/L)         160; 160         40/rep, 2 reps           Concentration 3 Nom; Meas (μg/L)         310; 310         40/rep, 2 reps           Concentration 4 Nom; Meas (μg/L)         630; 620         40/rep, 2 reps           Concentration 5 Nom; Meas (μg/L)         1300; 1200         40/rep, 2 reps           Concentration 6 Nom; Meas (μg/L)         2500; 2400         40/rep, 2 reps           Control         Negative: 0;         40/rep, 2 reps           NOEC         620         Method: Batlett's test or Scheffe's multiple comparison test p: 0.05		Acetone, 0.3 mL/L	
Concentration 2 Nom; Meas (μg/L)         160; 160         40/rep, 2 reps           Concentration 3 Nom; Meas (μg/L)         310; 310         40/rep, 2 reps           Concentration 4 Nom; Meas (μg/L)         630; 620         40/rep, 2 reps           Concentration 5 Nom; Meas (μg/L)         1300; 1200         40/rep, 2 reps           Concentration 6 Nom; Meas (μg/L)         2500; 2400         40/rep, 2 reps           Control         Negative: 0;         40/rep, 2 reps           Solvent: 0;         Method: Batlett's test or Scheffe's multiple comparison test p: 0.05			
Concentration 2 Nom; Meas (μg/L)         160; 160         40/rep, 2 reps           Concentration 3 Nom; Meas (μg/L)         310; 310         40/rep, 2 reps           Concentration 4 Nom; Meas (μg/L)         630; 620         40/rep, 2 reps           Concentration 5 Nom; Meas (μg/L)         1300; 1200         40/rep, 2 reps           Concentration 6 Nom; Meas (μg/L)         2500; 2400         40/rep, 2 reps           Control         Negative: 0; Solvent: 0;         40/rep, 2 reps           NOEC         620         Method: Batlett's test or Scheffe's multiple comparison test p: 0.05	Concentration 1 Nom; Meas (µg/L)	78; 81	40/rep, 2 reps
Concentration 3 Nom; Meas (μg/L)         310; 310         40/rep, 2 reps           Concentration 4 Nom; Meas (μg/L)         630; 620         40/rep, 2 reps           Concentration 5 Nom; Meas (μg/L)         1300; 1200         40/rep, 2 reps           Concentration 6 Nom; Meas (μg/L)         2500; 2400         40/rep, 2 reps           Control         Negative: 0;         40/rep, 2 reps           NOEC         620         Method: Batlett's test or Scheffe's multiple comparison test p: 0.05		160; 160	40/rep, 2 reps
Concentration 4 Nom; Meas (μg/L)         630; 620         40/rep, 2 reps           Concentration 5 Nom; Meas (μg/L)         1300; 1200         40/rep, 2 reps           Concentration 6 Nom; Meas (μg/L)         2500; 2400         40/rep, 2 reps           Control         Negative: 0;         40/rep, 2 reps           Solvent: 0;         Method: Batlett's test or Scheffe's multiple comparison test p: 0.05		-	
Concentration 5 Nom; Meas (μg/L)       1300; 1200       40/rep, 2 reps         Concentration 6 Nom; Meas (μg/L)       2500; 2400       40/rep, 2 reps         Control       Negative: 0; Solvent: 0;       40/rep, 2 reps         NOEC       620       Method: Batlett's test or Scheffe's multiple comparison test p: 0.05		<u>'</u>	* *
Concentration 6 Nom; Meas (μg/L)       2500; 2400       40/rep, 2 reps         Control       Negative: 0;       40/rep, 2 reps         Solvent: 0;       Method: Batlett's test or Scheffe's multiple comparison test p: 0.05		<u> </u>	1 1
Control  Negative: 0; Solvent: 0;  Method: Batlett's test or Scheffe's multiple comparison test p: 0.05		· ·	
NOEC  Solvent: 0;  Method: Batlett's test or Scheffe's multiple comparison test p: 0.05		*	
NOEC  620  Method: Batlett's test or Scheffe's multiple comparison test p: 0.05	Control		40/1cp, 2 1cps
test or Scheffe's multiple comparison test p: 0.05	NOFC		Method: Batlett's
multiple comparison test p: 0.05			
comparison test p: 0.05			
p: 0.05			-
			_
I VISID: not reported			MSD: not reported

	Graves et al. 1995	P. promelas
Parameter	Value	Comment
LOEC	1200	Method: Batlett's test or Scheffe's multiple comparison test
MATC (GeoMean NOEC, LOEC)	860	
% control at NOEC	Hatching success: 99 % Survival: 99 % Wet weight: 105 % Dry weight: 96 % Length: 99 %	Hatching success: 94 (tmt) / 94.5 (mean controls) = 99
		Survival: 95 (tmt) / 96 (mean controls) = 99
		Wet weight: 76.5 (tmt) / 72.9 (mean controls) = 105
		Dry weight: 15.8 (tmt) / 16.5 (mean controls) = 96
		Length: 18.8 (tmt) / 19 (mean controls) = 99
% control at LOEC	Hatching success: 102 % Survival: 101 % Wet weight: 93 % Dry weight: 83 % Length: 96 %	Hatching success: 96 (tmt) / 94.5 (mean controls) = 102
	Bengui. 90 %	Survival: 97 (tmt) / 96 (mean controls) = 101
		Wet weight: 67.8 (tmt) / 72.9 (mean controls) = 93
		Dry weight: 13.7 (tmt) / 16.5 (mean controls) = 83
		Length: 18.2 (tmt) / 19 (mean controls) = 96

Notes: Report notes that the solvent concentration used was higher than in all three testing guidelines but was done to enhance solubilization of the prometryn. The concentration of 0.30 mL/L was considered by the study authors to be within the tolerance of the fathead minnows for early life-stage toxicity testing. Points will not be deducted for the solvent concentration as a result.

Prometryn solubility (S) = 31,250 ug/L  $\mu$ g/L, 2S = 62,500  $\mu$ g/L.

Reliability points taken off for:

<u>Documentation:</u> Minimum significant difference (2), Point estimates (8). Total: 100-10 = 90 Acceptability: Minimum significant difference (1), Point estimates (3). Total: 100-4 = 96

Reliability score: mean(90, 96)=93

#### Pimephales promelas

Study: Humaker TL. (1985) Flow-through fathead minnow early life stage toxicity test with prometryn. Environmental Research and Technology, Inc., Fort Collins, CO. Laboratory study number D187. Ciba-Geigy Corporation, Greensboro, NC. USEPA MRID 40573721.

RelevanceReliabilityScore: 100Score: 86Rating: RRating: R

Relevance points taken off for: none

	Humaker 1985	P. promelas
Parameter	Value	Comment
Test method cited	ERT Bioassay Protocols D-	Appendix A
	5C and S-1C	
Phylum/subphylum	Chordata	
Class	Actinopterygii	
Order	Cypriniformes	
Family	Cyprinidae	
Genus	Pimephales	
Species	promelas	
Family native to North America?	Yes	
Age/size at start of test/growth	< 48 h embryos	
phase		
Source of organisms	Juveniles: Northeastern	
	Biologists Inc., Rhinebeck,	
	New York	
	Fertilized embryos: Sea	
	Plantations, Inc., Salem,	
	Massachusetts	
Have organisms been exposed to contaminants?	No	
Animals acclimated and disease-	Yes	
free?		
Animals randomized?	Yes	
Test vessels randomized?	Not reported	
Test duration	32 d	
Data for multiple times?	Yes	
Effect 1	Reproduction	% embryos producing live fry at end of test
Control response 1	Negative: 6. %2	
r	Solvent: 6.2 %	
Effect 2	Growth	
Control response 2	Length:	

	Humaker 1985	P. promelas
Parameter	Value	Comment
	Negative: 0.0965 g	
	Solvent: 0.0811 g	
	Weight:	
	Negative: 2.2523 mm	
	Solvent: 2.1593 g	
Temperature	20 ± 1 °C	
Test type	Flow-through chronic/early	
	life stage	
Photoperiod/light intensity	16l:8d	
Dilution water	Dechlorinated tap	
рН	6.9 - 7.5	
Hardness	20 - 28 mg/L CaCO <sub>3</sub>	
Alkalinity	18 - 30 mg/L	
Conductivity	90 - 135 umhos/cm	
Dissolved Oxygen	4.2 – 7.2 mg/L	46 – 79 %
Feeding	Live, < 48 h brine shrimp	
	2/d	
Purity of test substance	97 %	
Concentrations measured?	Yes	
Measured is what % of nominal?	84 - 116 %	
Toxicity values calculated based on	Measured	
nominal or measured		
concentrations?		
Chemical method documented?	GC	
Concentration of carrier (if any) in	Early life stage: 100 mg/L	
test solutions	(0.13  mL/L) for all	
	treatments	
Concentration 1 Nom; Meas (µg/L)	310; 356	34/rep, 2 reps
Concentration 2 Nom; Meas (µg/L)	610; 802	34/rep, 2 reps
Concentration 3 Nom; Meas (µg/L)	1200; 1390	34/rep, 2 reps
Concentration 4 Nom; Meas (µg/L)	2520; 2582	34/rep, 2 reps
Concentration 5 Nom; Meas (µg/L)	4920; 4148	34/rep, 2 reps
Control	Solvent: 0; 0	34/rep, 2 reps
	Negative: 0; 0	
	802 μg/L	Method: moving
NOEC		average
		p: 0.05
		MSD: n/a
LOEC	1390 μg/L	
MATC (GeoMean NOEC, LOEC)	1056 μg/L	
% control at NOEC	Length: 94	NOEC: 802
	Weight: 93	Length: 2.0763
		(tmt) / 2.2058
		(mean controls) =

	Humaker 1985	P. promelas
Parameter	Value	Comment
		94
		Weight: 0.0826
		(tmt) / 0.0888
		(mean controls) =
		93
% control at LOEC	Length: 91	NOEC: 1390
	Weight: 89	Length: 2.0026
		(tmt) / 2.2058
		(mean controls) =
		91
		Weight: 0.0792
		(tmt) / 0.0888
		(mean controls) =
		89

Prometryn solubility (S) = 31,250 ug/L  $\mu$ g/L, 2S = 62,500  $\mu$ g/L. All exposure concentrations acceptable.

Reliability points taken off for:

<u>Documentation:</u> Minimum significant difference (2), Point estimates (8). Total: 100 - 10 = 90

<u>Acceptability:</u> Carrier solvent (4), Dissolved oxygen (6), Minimum significant difference (1), Point estimates (3). Total: 100 - 14 = 86

Reliability score: mean (90, 86) = 88

#### Raphidocelis subcapitata

Study: Hughes JS. (1987). The toxicity of prometryn technical to *Selanstrum capricornutum* EPA Guidelines No. 123-2. Conducted by Malcolm Pirnie, Inc., White Plains, New York. Laboratory study number 0267-38-1100-1. Presented to Ciba-Geigy Corporation, Agricultural Division, Greensboro, North Carolina. CADPR study ID 140027.

RelevanceReliabilityScore: 100Score: 94.5Rating: RRating: R

	Hughes 1987	R. subcapitata
Parameter	Value	Comment
Test method cited	USEPA-FIFRA Pesticide	
	Assessment Guidelines	
	Subdivision J, Hazard	
	Evaluation: Non-Target	
	Plants Guideline 123-2,	
	Growth and Reproduction	
	of Aquatic Plants, Tier 2	
Phylum/subphylum	Chlorophyta	
Class	Chlorophyceae	
Order	Sphaeropleales	
Family	Selenastraceae	
Genus	Raphidocelis	
Species	subcapitata	
Family native to North America?	Yes	
Age/size at start of test/growth	7 d	
phase		
Source of organisms	Laboratory stock cultures	
Have organisms been exposed to	No	
contaminants?		
Animals acclimated and disease-	Yes	
free?		
Animals randomized?	Yes	Aliquots
Test vessels randomized?	Yes	
Test duration	7 d	
Data for multiple times?	Yes	2, 3, 4, 7
Effect 1	Cell count	
Control response 1	2 d: 43,667	
	3 d: 352,333	
	4 d: 1,859,333	
	7 d: 10,226,667	
Temperature	$24 \pm 2$ °C	
Test type	Static	
Photoperiod/light intensity	Continuous/ $4306 \pm 650$ lux	

	Hughes 1987	R. subcapitata
Parameter	Value	Comment
Dilution water	Growth medium made with	Synthetic algal
	DI water	assay procedure
		nutrient medium;
		ingredients in App.
		В
рН	$7.5 \pm 0.1$	
Hardness	Not reported but used DI	
	water	
Alkalinity	Not reported but used DI	
	water	
Conductivity	Not reported but used DI	
	water	
Dissolved Oxygen	Not reported but used DI	
	water	
Feeding	Growth medium	
Purity of test substance	98.1 %	
Concentrations measured?	Yes	
Measured is what % of nominal?	73-93 %	
Toxicity values calculated based on	Measured	
nominal or measured		
concentrations?		
Chemical method documented?	GC-ECD	Performed by
		outside company:
		EN-CAS
		laboratories,
		Winston-Salem,
		North Carolina
Concentration of carrier (if any) in	None used	
test solutions	5.26	2 150,000
Concentration 1 Nom; Meas (µg/L)	5; 3.6	3 reps, 150,000
	10.01	cells/rep
Concentration 2 Nom; Meas (µg/L)	10; 8.1	3 reps, 150,000
	20.16	cells/rep
Concentration 3 Nom; Meas (µg/L)	20; 16	3 reps, 150,000
	40. 26	cells/rep
Concentration 4 Nom; Meas (µg/L)	40; 36	3 reps, 150,000
	00.62	cells/rep
Concentration 5 Nom; Meas (µg/L)	80; 62	3 reps, 150,000
C41	0 5	cells/rep
Control	0; < 5	3 reps, 150,000
EC (050) CD ( // // )	22*	cells/rep
EC <sub>50</sub> (95% CI) (μg/L)	23*	Method: probit
NOEC	8.1	Method: ANOVA
		and multiple range
		tests
		p: 0.05

	Hughes 1987	R. subcapitata
Parameter	Value	Comment
		MSD: NR
LOEC	16	
MATC	11	
% control at NOEC	7 d: 110 %	NOEC = 8.1 7 d: 11,240,000 (tmt) / 10,226,667 (control) = 110 %
% control at LOEC	7 d: 88 %	LOEC = 16 7 d: 8,960,000 (tmt) / 10,226,667 (control) = 88 %

Notes: Growth medium prepared by standard recipe and used distilled water. Therefore, dilution water parameters were not reported but are considered adequate for the test species.

Prometryn solubility (S) = 31,250 ug/L  $\mu$ g/L, 2S = 62,500  $\mu$ g/L. All exposure concentrations were acceptable.

\*Confidence limits could not be calculated because "an error condition arises in the calculations as a result of an attempt to take the square root of a negative number."

Reliability points taken off for:

<u>Documentation</u>: Statistical significance (2), Significance level (2), Minimum significant difference (2). Total: 100 - 6 = 94

<u>Acceptability:</u> Temperature variation (3), Minimum significant difference (1), % control at LOEC (1). Total: 100 - 5 = 95

Reliability score: mean (94, 95) = 94.5

Appendix A2 - Wildlife Toxicity Studies Rated R

Anas platyrhynchos

Fletcher et al. 1989. CADPR study ID 139999. EPA MRID 41035901.

Notes: Chronic reproduction study. No point estimates reported. No statistical significance observed between any exposure concentration and control.

Parameter <sup>1</sup>	Score <sup>2</sup>	Points
Exposure duration	20	20
Control type	7	7
Organism information (i.e., age, life stage)	8	8
Chemical grade or purity	5	5
Chemical analysis method	5	0
Exposure type (i.e., dermal, dietary, gavage)	10	10
Test location (i.e., laboratory, field, natural artificial)	5	5
Application frequency	5	5
Organism source	5	5
Organism number and/or sample number	5	5
Dose number	5	5
Statistics		
Hypothesis tests		
Statistical significance	5	5
Significance level	5	5
Minimum significant difference	3	0
% of control at NOEC and/or LOEC	3	0
Point estimates (i.e., LC <sub>50</sub> , EC <sub>50</sub> )	4	0
Total	100	85

<sup>&</sup>lt;sup>1</sup> Compiled from RIVM (2001), USEPA (1985; 2003b), ECOTOX (2006), CCME (1999), ANZECC & ARMCANZ (2000), OECD (1995), and Van Der Hoeven *et al.* (1997).

<sup>&</sup>lt;sup>2</sup> Weighting based acceptability criteria from various ASTM, OECD, APHA, and USEPA methods, ECOTOX (2006), and on data quality criteria in RIVM (2001), USEPA (1985; 2003b), CCME (1999), ANZECC & ARMCANZ (2000), OECD (1995), and Van Der Hoeven *et al.* (1997).

*Anas platyrhynchos*Beavers & Fink 1977. USEPA MRID 82966.

Notes: 8 day duration, technical grade,  $LC_{50} > 4640$  mg/kg

Parameter <sup>1</sup>	Score <sup>2</sup>	Points
Exposure duration	20	20
Control type	7	7
Organism information (i.e., age, life stage)	8	8
Chemical grade or purity	5	5
Chemical analysis method	5	0
Exposure type (i.e., dermal, dietary, gavage)	10	10
Test location (i.e., laboratory, field, natural artificial)	5	5
Application frequency	5	5
Organism source	5	5
Organism number and/or sample number	5	5
Dose number	5	5
Statistics		
Hypothesis tests		
Statistical significance	5	0
Significance level	5	5
Minimum significant difference	3	0
% of control at NOEC and/or LOEC	3	0
Point estimates (i.e., LC <sub>50</sub> , EC <sub>50</sub> )	4	4
Total	100	84

<sup>&</sup>lt;sup>1</sup> Compiled from RIVM (2001), USEPA (1985; 2003b), ECOTOX (2006), CCME (1999), ANZECC & ARMCANZ (2000), OECD (1995), and Van Der Hoeven *et al.* (1997).

<sup>&</sup>lt;sup>2</sup> Weighting based acceptability criteria from various ASTM, OECD, APHA, and USEPA methods, ECOTOX (2006), and on data quality criteria in RIVM (2001), USEPA (1985; 2003b), CCME (1999), ANZECC & ARMCANZ (2000), OECD (1995), and Van Der Hoeven *et al.* (1997).

*Anas platyrhynchos* Fletcher, D. 1984. CADPR study ID 139998.

Notes: 8 day duration, technical grade,  $LC_{50} > 5000$  mg/kg

Parameter <sup>1</sup>	Score <sup>2</sup>	Points
Exposure duration	20	20
Control type	7	7
Organism information (i.e., age, life stage)	8	8
Chemical grade or purity	5	5
Chemical analysis method	5	5
Exposure type (i.e., dermal, dietary, gavage)	10	10
Test location (i.e., laboratory, field, natural artificial)	5	5
Application frequency	5	5
Organism source	5	5
Organism number and/or sample number	5	5
Dose number	5	5
Statistics		
Hypothesis tests		
Statistical significance	5	0
Significance level	5	0
Minimum significant difference	3	0
% of control at NOEC and/or LOEC	3	0
Point estimates (i.e., LC <sub>50</sub> , EC <sub>50</sub> )	4	4
Total	100	84

<sup>&</sup>lt;sup>1</sup> Compiled from RIVM (2001), USEPA (1985; 2003b), ECOTOX (2006), CCME (1999), ANZECC & ARMCANZ (2000), OECD (1995), and Van Der Hoeven *et al.* (1997).

<sup>&</sup>lt;sup>2</sup> Weighting based acceptability criteria from various ASTM, OECD, APHA, and USEPA methods, ECOTOX (2006), and on data quality criteria in RIVM (2001), USEPA (1985; 2003b), CCME (1999), ANZECC & ARMCANZ (2000), OECD (1995), and Van Der Hoeven *et al.* (1997).

### Anas platyrhynchos

Study: Fletcher DW, Pedersen, CA. (1988) Prometryn technical: 28-day dietary toxicity and reproduction pilot study in mallard ducks. Bio-Life Associates, Limited, Neillsville, Wisconsin. Laboratory study number 87 DRP 21. Submitted to Ciba-Geigy Corporation, Greensboro, NC. USEPA MRID 43370402.

Notes: Pilot study. No point estimates determined. No adverse effects at any tested concentration.  $EC_{50}$  can be stated as >1,000 mg/kg.

Parameter <sup>1</sup>	Score <sup>2</sup>	Points
Exposure duration	20	20
Control type	7	7
Organism information (i.e., age, life stage)	8	8
Chemical grade or purity	5	5
Chemical analysis method	5	0
Exposure type (i.e., dermal, dietary, gavage)	10	10
Test location (i.e., laboratory, field, natural artificial)	5	5
Application frequency	5	5
Organism source	5	5
Organism number and/or sample number	5	5
Dose number	5	5
Statistics		
Hypothesis tests		
Statistical significance	5	0
Significance level	5	0
Minimum significant difference	3	0
% of control at NOEC and/or LOEC	3	0
Point estimates (i.e., LC <sub>50</sub> , EC <sub>50</sub> )	4	0
Total	100	80

<sup>&</sup>lt;sup>T</sup>Compiled from RIVM (2001), USEPA (1985; 2003b), ECOTOX (2006), CCME (1999), ANZECC & ARMCANZ (2000), OECD (1995), and Van Der Hoeven *et al.* (1997).

<sup>&</sup>lt;sup>2</sup> Weighting based acceptability criteria from various ASTM, OECD, APHA, and USEPA methods, ECOTOX (2006), and on data quality criteria in RIVM (2001), USEPA (1985; 2003b), CCME (1999), ANZECC & ARMCANZ (2000), OECD (1995), and Van Der Hoeven *et al.* (1997).

# Appendix A3 - Mesocosm studies rated R

Prometryn. Leibig et al. 2008.

Include notes next to each parameter.

Documentation and acceptability (reliability) evaluation for data derived from aquatic outdoor field and indoor model ecosystems experiments. Adapted from ECOTOX 2006; Table from TenBrook et al. 2010.

Parameter <sup>a</sup>	Score <sup>b</sup>	Points
Results published or in signed, dated format Published peer review article	5	5
Exposure duration and sample regime adequately described	6	6
Unimpacted site (Score 7 for artificial systems) artificial system	7	7
Adequate range of organisms in system (1° producers, 1°, 2° consumers) <b>Producers:</b> <i>Cryptomonas</i> sp., consumers: <i>Urotricha furcata</i> , decomposers: unspecified bacteria	6	6
Chemical		
Grade or purity stated 99.2	6	6
Concentrations measured/estimated and reported Nominal and measured reported	8	8
Analysis method stated GC-MS	2	2
Habitat described (e.g., pond, lake, ditch, artificial, lentic, lotic) laboratory system, lake model	6	6
Water quality		
Source identified Growth medium	2	2
Hardness reported No	1	0
Alkalinity reported <b>No</b>	1	0
Dissolved oxygen reported > 90%	2	2
Temperature reported 20 ± 1.5 °C	2	2
Conductivity reported No	1	0
pH reported $7.15 \pm 0.35$	1	0
Photoperiod reported Continuous/33 $\pm$ 3 $\mu$ mol photons m <sup>-2</sup> s <sup>-1</sup>	1	1
Organic carbon reported Table 2	2	2
Chemical fate reported	3	0
Geographic location identified (Score 2 for indoor systems) Indoor systems	2	2
Pesticide application		
Type reported (e.g., spray, dilutor, injection) Growth medium	2	2
Frequency reported Static	2	2
Date/season reported (Score 2 for indoor systems) Indoor system	2	2

Parameter <sup>a</sup>	Score <sup>b</sup>	Points
Test endpoints		
Species abundance reported cell number of producers/consumers, graphically	3	3
Species diversity reported	3	0
Biomass reported only TOC/TN as concentrations	2	0
Ecosystem recovery reported	2	0
Statistics		
Methods identified ANOVA, probit, Student t-test with Bonferroni adjustment	2	2
At least 2 replicates 4 reps/treatment, 6 reps/control	3	3
At least 2 test concentrations and 1 control 3 conc. & control	3	3
Dose-response relationship observed	2	2
Hypothesis tests		
NOEC determined	4	4
Significance level stated $\alpha = 0.05$	2	2
Minimum significant difference reported	2	0
% of control at NOEC and/or LOEC reported or calculable <b>control values not tabulated, only plotted</b>	2	0
Total Reliability	100	82

LOEC = lowest observed effect concentration, NOEC = no observed effect concentration.

<sup>a</sup>Compiled from RIVM 2001, USEPA 1985 and 2003a, ECOTOX 2006, CCME 1995, ANZECC and ARMCANZ 2000, OECD 1995a, and van der Hoeven et al. 1997.

<sup>b</sup>Weighting based on ECOTOX 2006 and on data quality criteria in RIVM 2001 and OECD 1995a.

# Appendix A4 – Studies rated RL, LR, LL

### Americamysis bahia

Study: Claude MB, Kendall TZ, Krueger HO. (2013) Prometryn-A flow-through life-cycle toxicity test with the saltwater mysid (*Americamysis bahia*). Wildlife International, Easton, MD study number 528A-239A. Submitted to Syngenta Crop Protection, LLC, Greensboro, NC. CADPR study ID 490745-01.

RelevanceReliabilityScore: 85Score: 86Rating: LRating: R

Relevance points taken off for: Freshwater (15). 100 - 15 = 85

	Claude et al. 2013	A. bahia
Parameter	Value	Comment
Test method cited	OPPTS 580.10350 and	
	ASTM Standard E 1191-03a	
Phylum/subphylum	Arthropoda/Crustacea	
Class	Malacostraca	
Order	Mysida	
Family	Mysidae	
Genus	Americamysis	
Species	bahia	
Family native to North America?	Yes	
Age/size at start of test/growth	Neonates (< 24 h)	
phase		
Source of organisms	Wildlife International	
	laboratory cultures, Easton,	
	Maryland	
Have organisms been exposed to	No	
contaminants?		
Animals acclimated and disease-	Yes	14 d
free?		
Animals randomized?	Yes	
Test vessels randomized?	Yes	
Test duration	28 d	
Data for multiple times?	No	
Effect 1	Adult survival	
Control response 1	Negative: 96 %	Statistical
	Solvent: 83.3 %	difference between
		controls $(p \le 0.05)$
Effect 2	Reproduction (number of young)	
Control response 2	Negative: 100 %	No statistical
•	Solvent: 94.4 %	difference so
		controls pooled

Parameter         Value         Comment           Effect 3         Growth         No statistical difference so controls pooled           Control response 3         Negative: % solvent: % difference so controls pooled           Temperature $25 \pm 2^{\circ}$ C           Test type         Flow-through         120 m transition period           Photoperiod/light intensity         14 1:10 d; 195 lux         120 m transition period           Dilution water         Natural seawater, filtered, UV-sterilized diluted with on-site well water         Indian River Inlet, Delaware           pH         8.1 ± 0.1         Hardness           Alkalinity         Not reported         Delaware           Conductivity         Not reported         Not reported           Dissolved Oxygen         ≥ 7.1, ≥ 97 %         INVE Aquaculture, Salt Lake City, Utah           Feeding         Live brine shrimp (Artemia sp.), daily and periodically suppl. with nutrient enrichment (AI DHA Selco from InVF Thailand, Ltd.) and Skeletonema costatum (Wildlife International)         INVE Aquaculture, Salt Lake City, Utah           Purity of test substance         97.8 %         Inversion of Concentrations measured?         Yes           Measured         Measured         Inversion of Concentration of Carrier (if any) in test solutions         Inversion of Concentration of Carrier (if any) in test solutions         Inversion of Concentration of		Claude et al. 2013	A. bahia
Control response 3       Negative: % Solvent: %       No statistical difference so controls pooled         Temperature $25 \pm 2$ °C         Test type       Flow-through         Photoperiod/light intensity $14 \text{ l: 10 d; 195 lux}$ $120 \text{ m transition period}$ Dilution water       Natural seawater, filtered, UV-sterilized diluted with on-site well water       Indian River Inlet, Delaware         pH $8.1 \pm 0.1$ Hardness         Alkalinity       Not reported       Onductivity         Conductivity       Not reported       Indian River Inlet, Delaware         Conductivity       Not reported       Indian River Inlet, Delaware         Dissolved Oxygen $\geq 7.1, \geq 97$ %       INVE Aquaculture, Salt Lake City, Utah         Feeding       Live brine shrimp (Artemia sp.), daily and periodically suppl. with nutrient enrichment (A1 DHA Selco from INVE Thailand, Ltd.) and Skeletonema costatum (Wildlife International)       INVE Aquaculture, Salt Lake City, Utah         Purity of test substance       97.8 %       Measured         Concentrations measured?       Yes         Measured is what % of nominal?       84-90         Toxicity values calculated based on nominal or measured       Measured         Concentration of carrier (if any) in test solutions       100 method documented?         Concentration 1 Nom; Meas (µg	Parameter	Value	Comment
Solvent: %   difference so controls pooled	Effect 3	Growth	
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$	Control response 3		No statistical
Temperature Test type Flow-through Photoperiod/light intensity Flow-through Photoperiod/light intensity Flat:10 d; 195 lux Flow-through Flow-throug		Solvent: %	
Test type Photoperiod/light intensity 14 1:10 d; 195 lux 120 m transition period Dilution water Natural seawater, filtered, UV-sterilized diluted with on-site well water 8.1 ± 0.1 Hardness Not reported Alkalinity Not reported Dissolved Oxygen 2.1, ≥ 97 % Live brine shrimp (Artemia sp.), daily and periodically suppl. with nutrient enrichment (A1 DHA Selco from INVE Thailand, Ltd.) and Skeletonema costatum (Wildlife International) Purity of test substance 97.8 % Concentrations measured? Yes Measured is what % of nominal? 84-90 Measured on nominal or measured concentrations? Chemical method documented? HPLC/UV Concentration of carrier (if any) in test solutions HPLC/UV dimethylformamide Concentration 1 Nom; Meas (μg/L) Concentration 2 Nom; Meas (μg/L) Concentration 3 Nom; Meas (μg/L) Concentration 5 Nom; Meas (μg/L) Concentration 6 Nom; Meas (μg/L) Concentration 7 Nom; Meas (μg/L) Concentration 6 Nom; Meas (μg/L)			controls pooled
Photoperiod/light intensity       14 1:10 d; 195 lux       120 m transition period         Dilution water       Natural seawater, filtered, UV-sterilized diluted with on-site well water       Indian River Inlet, Delaware         pH       8.1 ± 0.1       Hardness         Alkalinity       Not reported       Alkalinity         Conductivity       Not reported       Dissolved Oxygen         Feeding       Live brine shrimp (Artemia sp.), daily and periodically suppl. with nutrient enrichment (A1 DHA Selco from INVE Thailand, Ltd.) and skeletonema costatum (Wildlife International)       Salt Lake City, Utah         Purity of test substance       97.8 %       Concentrations measured?         Measured is what % of nominal?       84-90         Toxicity values calculated based on nominal or measured concentrations?       Measured         Chemical method documented?       HPLC/UV         Concentration of carrier (if any) in test solutions       HPLC/UV         Concentration 1 Nom; Meas (µg/L)       0.063; 0.055       4 reps, 15/rep first generation 4 reps, 5 mating pairs/rep at sexual maturity         Concentration 2 Nom; Meas (µg/L)       250; 220         Concentration 5 Nom; Meas (µg/L)       500;450         Concentration 5 Nom; Meas (µg/L)       1000; 840         Control; negative, solvent (µg/L)       0; < LOQ	Temperature	25 ± 2 °C	
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$		ŭ	
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$	Photoperiod/light intensity	14 l:10 d; 195 lux	
On-site well water   PH   8.1 ± 0.1	Dilution water		*
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$			Delaware
Hardness       Not reported         Alkalinity       Not reported         Conductivity       Not reported         Dissolved Oxygen $\geq 7.1, \geq 97\%$ Feeding       Live brine shrimp (Artemia sp.), daily and periodically suppl. with nutrient enrichment (A1 DHA Selco from INVE Thailand, Ltd.) and Skeletonema costatum (Wildlife International)       Salt Lake City, Utah         Purity of test substance       97.8 %       Utah         Concentrations measured?       Yes         Measured is what % of nominal?       84-90         Toxicity values calculated based on nominal or measured concentrations?       Measured         Chemical method documented?       HPLC/UV         Concentration of carrier (if any) in test solutions       Unimethylformamide         Concentration 1 Nom; Meas (µg/L)       0.063; 0.055       4 reps, 15/rep first generation 4 reps, 5 mating pairs/rep at sexual maturity         Concentration 2 Nom; Meas (µg/L)       250; 220         Concentration 5 Nom; Meas (µg/L)       500; 450         Concentration 5 Nom; Meas (µg/L)       1000; 840         Control; negative, solvent (µg/L)       0; < LOQ			
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$		$8.1 \pm 0.1$	
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$		1	
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$	·	*	
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$			
sp.), daily and periodically suppl. with nutrient enrichment (A1 DHA Selco from INVE Thailand, Ltd.) and Skeletonema costatum (Wildlife International)       Salt Lake City, Utah         Purity of test substance       97.8 %         Concentrations measured?       Yes         Measured is what % of nominal?       84-90         Toxicity values calculated based on nominal or measured concentrations?       Measured         Chemical method documented?       HPLC/UV         Concentration of carrier (if any) in test solutions       dimethylformamide         Concentration 1 Nom; Meas (μg/L)       0.063; 0.055       4 reps, 15/rep first generation 4 reps, 5 mating pairs/rep at sexual maturity         Concentration 2 Nom; Meas (μg/L)       130; 110       Concentration 3 Nom; Meas (μg/L)         Concentration 4 Nom; Meas (μg/L)       500;450         Concentration 5 Nom; Meas (μg/L)       1000; 840         Control; negative, solvent (μg/L)       0; < LOQ		-	
$\begin{array}{c} \text{suppl. with nutrient} \\ \text{enrichment (A1 DHA Selco} \\ \text{from INVE Thailand, Ltd.)} \\ \text{and $Skeletonema \ costatum} \\ \text{(Wildlife International)} \\ \end{array}$	Feeding	<b>.</b> `	_
enrichment (A1 DHA Selco from INVE Thailand, Ltd.) and Skeletonema costatum (Wildlife International)  Purity of test substance 97.8 %  Concentrations measured? Yes  Measured is what % of nominal? 84-90  Toxicity values calculated based on nominal or measured concentrations?  Chemical method documented? HPLC/UV  Concentration of carrier (if any) in test solutions dimethylformamide  Concentration 1 Nom; Meas ( $\mu$ g/L) 0.063; 0.055 4 reps, 15/rep first generation 4 reps, 5 mating pairs/rep at sexual maturity  Concentration 2 Nom; Meas ( $\mu$ g/L) 130; 110  Concentration 3 Nom; Meas ( $\mu$ g/L) 250; 220  Concentration 4 Nom; Meas ( $\mu$ g/L) 500;450  Concentration 5 Nom; Meas ( $\mu$ g/L) 1000; 840  Control; negative, solvent ( $\mu$ g/L) 0; < LOQ Negative: dilution water Solvent: 0.02 mL/L			,
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$			Utah
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$		,	
(Wildlife International)         Purity of test substance       97.8 %         Concentrations measured?       Yes         Measured is what % of nominal?       84-90         Toxicity values calculated based on nominal or measured concentrations?       Measured         Chemical method documented?       HPLC/UV         Concentration of carrier (if any) in test solutions       0.02 mL/L dimethylformamide         Concentration 1 Nom; Meas (μg/L)       0.063; 0.055       4 reps, 15/rep first generation 4 reps, 5 mating pairs/rep at sexual maturity         Concentration 2 Nom; Meas (μg/L)       130; 110         Concentration 3 Nom; Meas (μg/L)       250; 220         Concentration 4 Nom; Meas (μg/L)       500;450         Concentration 5 Nom; Meas (μg/L)       1000; 840         Control; negative, solvent (μg/L)       0; < LOQ			
$ \begin{array}{c ccccccccccccccccccccccccccccccccccc$			
Measured is what % of nominal?84-90Toxicity values calculated based on nominal or measured concentrations?MeasuredChemical method documented?HPLC/UVConcentration of carrier (if any) in test solutions0.02 mL/L dimethylformamideConcentration 1 Nom; Meas ( $\mu$ g/L)0.063; 0.0554 reps, 15/rep first generation 4 reps, 5 mating pairs/rep at sexual maturityConcentration 2 Nom; Meas ( $\mu$ g/L)130; 110Concentration 3 Nom; Meas ( $\mu$ g/L)250; 220Concentration 4 Nom; Meas ( $\mu$ g/L)500;450Concentration 5 Nom; Meas ( $\mu$ g/L)1000; 840Control; negative, solvent ( $\mu$ g/L)0; < LOQ	•		
$\begin{array}{c} nominal \ or \ measured \\ concentrations? \\ \hline Chemical \ method \ documented? \\ \hline Concentration \ of \ carrier \ (if \ any) \ in \\ test \ solutions \\ \hline Concentration \ 1 \ Nom; \ Meas \ (\mu g/L) \\ \hline Concentration \ 2 \ Nom; \ Meas \ (\mu g/L) \\ \hline Concentration \ 2 \ Nom; \ Meas \ (\mu g/L) \\ \hline Concentration \ 3 \ Nom; \ Meas \ (\mu g/L) \\ \hline Concentration \ 4 \ Nom; \ Meas \ (\mu g/L) \\ \hline Concentration \ 5 \ Nom; \ Meas \ (\mu g/L) \\ \hline Concentration \ 5 \ Nom; \ Meas \ (\mu g/L) \\ \hline Concentration \ 5 \ Nom; \ Meas \ (\mu g/L) \\ \hline Control; \ negative, \ solvent \ (\mu g/L) \\ \hline Control; \ negative, \ solvent \ (\mu g/L) \\ \hline \end{array}$			
		Measured	
$ \begin{array}{c} \hbox{Chemical method documented?} & \hbox{HPLC/UV} \\ \hbox{Concentration of carrier (if any) in test solutions} & \hbox{0.02 mL/L} \\ \hbox{Concentration 1 Nom; Meas ($\mu g/L$)} & \hbox{0.063; 0.055} & \hbox{4 reps, 15/rep first generation} \\ \hbox{Concentration 2 Nom; Meas ($\mu g/L$)} & \hbox{130; 110} \\ \hbox{Concentration 3 Nom; Meas ($\mu g/L$)} & \hbox{250; 220} \\ \hbox{Concentration 4 Nom; Meas ($\mu g/L$)} & \hbox{500;450} \\ \hbox{Concentration 5 Nom; Meas ($\mu g/L$)} & \hbox{1000; 840} \\ \hbox{Control; negative, solvent ($\mu g/L$)} & \hbox{0; < LOQ} & \hbox{Negative: dilution water} \\ \hbox{Solvent: 0.02 mL/L} \\ \end{array} $			
$ \begin{array}{c} \mbox{Concentration of carrier (if any) in test solutions} & 0.02 \mbox{ mL/L} \\ \mbox{dimethylformamide} & 4 \mbox{ reps, 15/rep first generation} \\ \mbox{Concentration 1 Nom; Meas (\mu g/L)} & 0.063; 0.055 & 4 \mathref{reps, 5 mating pairs/rep at sexual maturity}} \\ \mbox{Concentration 2 Nom; Meas (\mu g/L)} & 130; 110 & 250; 220 & 250; 220 & 250; 220 & 250; 220 & 250; 250; 220 & 250; 250; 250; 250; 250; 250; 250; 250;$		HDI CANA	
$\begin{array}{c} \text{test solutions} & \text{dimethylformamide} \\ \text{Concentration 1 Nom; Meas ($\mu g/L$)} & 0.063; 0.055 & 4 \text{ reps, 15/rep first} \\ & & & & & & & & & & & \\ & & & & & & $			
$ \begin{array}{c} \text{Concentration 1 Nom; Meas ($\mu g/L$)} & 0.063;  0.055 \\ & & 4 \text{ reps, } 15 \text{/rep first} \\ & & \text{generation} \\ & 4 \text{ reps, } 5 \text{ mating} \\ & \text{pairs/rep at sexual} \\ & \text{maturity} \\ \hline \\ \text{Concentration 2 Nom; Meas ($\mu g/L$)} & 130;  110 \\ \hline \\ \text{Concentration 3 Nom; Meas ($\mu g/L$)} & 250;  220 \\ \hline \\ \text{Concentration 4 Nom; Meas ($\mu g/L$)} & 500; \! 450 \\ \hline \\ \text{Concentration 5 Nom; Meas ($\mu g/L$)} & 1000;  840 \\ \hline \\ \text{Control; negative, solvent ($\mu g/L$)} & 0; < LOQ \\ \hline \\ \text{Negative: dilution} \\ & \text{water} \\ & \text{Solvent: } 0.02 \text{ mL/L} \\ \hline \end{array} $			
$\begin{array}{c} & & & & & & \\ & & & & & \\ & & & & & \\ & & & & \\ & & & & \\ & & & & \\ & & & \\ & & & \\ & & & \\ & & & \\ &$		·	A none 15/non finat
	Concentration I Nom; Meas (µg/L)	0.063; 0.055	
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$			_
			_
			•
	Concentration 2 Nom; Meas (µg/L)	130; 110	
		250; 220	
$ \begin{array}{c cccc} Concentration 5 & Nom; & Meas & (\mu g/L) & 1000; & 840 \\ Control; & negative, & solvent & (\mu g/L) & 0; & < LOQ & & Negative: & dilution & water & Solvent: & 0.02 & mL/L \\ \hline \end{array} $			
Control; negative, solvent ( $\mu g/L$ ) 0; < LOQ Negative: dilution water Solvent: 0.02 mL/L		,	
water Solvent: 0.02 mL/L	•		Negative: dilution
Solvent: 0.02 mL/L	Control, negative, solvent (µg/L)	o, .20 Q	_
NOEC Adult survival: 450 μg/L Method:	NOEC	Adult survival: 450 ug/L	•

	Claude et al. 2013	A. bahia
Parameter	Value	Comment
	Reproduction: 110 μg/L Growth: 450 μg/L	Wilcoxon's rank sum with Bonferroni adjustment p: 0.05 MSD: n/a
LOEC	Adult survival: 840 μg/L Reproduction: 220 μg/L Growth: 840 μg/L	
MATC (GeoMean NOEC, LOEC)	Adult survival: 615 Reproduction: 1600 Growth: 615	
% control at NOEC	Adult survival: 92.3 Reproduction: 9.3 Length: 98.6 Weight: 93.6	Adult survival (NOEC = 450): 90/97.5 = 92.3 %  Reproduction (NOEC = 840): 9.1/97.4 = 9.3  Growth (NOEC = 450): Length (mean M/F): mean(7.83, 7.96) / mean(7.97, 8.05) = 98.6  Weight: (mean M/F): mean(0.76,0.99) / mean(1.06,0.81) = 93.6
% control at LOEC	Adult survival: 87.1 Reproduction: 89 Length: 91.6 Weight: 72.2	Adult survival (LOEC = 840): 85/97.5 = 87.1  Reproduction (LOEC = 220): 86.7/97.4 = 89  Growth (LOEC = 840): Length (mean M/F): mean(7.43,7.24) / mean(7.97, 8.05) = 91.6

	Claude et al. 2013	A. bahia
Parameter	Value	Comment
		Weight: (mean M/F): mean(0.69, 0.66) / mean(1.06,0.81) = 72.2

Notes: Saltwater species tested in brackish water with salinity of 19-20 %.

Prometryn solubility (S) = 31,250 ug/L  $\mu$ g/L, 2S = 62,500  $\mu$ g/L. All exposure concentrations were acceptable.

Reliability points taken off for:

<u>Documentation:</u> Hardness (2), Alkalinity (2), Conductivity (2), Minimum significant difference (2), Point estimates (8). Total: 100 - 16 = 84

<u>Acceptability:</u> Hardness (2), Alkalinity (2), Temperature variation (3), Conductivity (1), Minimum significant difference (1), Point estimates (3). Total: 100 - 12 = 88

Reliability score: mean (84, 88) = 86

#### Americamysis bahia

Study: Surprenant DC. (1988a) Acute toxicity of prometryn technical to mysid shrimp (*Mysidopsis bahia*). Springborn Life Sciences, Inc., Wareham, MA. Laboratory study number 88-1-2601. Ciba-Geigy Corporation, Greensboro, NC. USEPA MRID 40573718.

RelevanceReliabilityScore: 85Score: 85.5Rating: LRating: R

Relevance points taken off for: Freshwater (15). 100 - 15 = 85.

	Surprenant 1988a	A. bahia
Parameter	Value	Comment
Test method cited	Protocol for Conducting a	
	Static Acute Toxicity Test	
	with Mysidopsis bahia,	
	Following FIFRA	
	Guidelines, SLS Protocol #	
	120387/MYS.SA	
Phylum/subphylum	Arthropoda/Crustacea	
Class	Malacostraca	
Order	Mysida	
Family	Mysidae	
Genus	Americamysis	
Species	bahia	
Family native to North America?	Yes	
Age/size at start of test/growth	< 24 h	
phase		
Source of organisms	SLS culture facility	
Have organisms been exposed to	No	
contaminants?		
Animals acclimated and disease-	Yes	
free?		
Animals randomized?	Yes	
Test vessels randomized?	Not reported	
Test duration	96 h	
Data for multiple times?	24, 48, 72, 96 h	
Effect 1	Mortality	
Control response 1	2.5 %	97.5% survival
Temperature	25 ± 1 °C	
Test type	Static	
Photoperiod/light intensity	16 l: 8 d/50-90 footcandles	
Dilution water	Natural filtered seawater	Salinity: 32 ‰
	from Cape Cod Canal,	
	Bourne, Massachusetts	

	Surprenant 1988a	A. bahia
Parameter	Value	Comment
pH	7.9	
Hardness	Not reported	
Alkalinity	Not reported	
Conductivity	Not reported	
Dissolved Oxygen	5.9 mg/L	71.4%
Feeding	Live brine shrimp nauplii	
	daily	
Purity of test substance	98.1 %	
Concentrations measured?	Yes	
Measured is what % of nominal?	60-106%	
Toxicity values calculated based on	Measured	
nominal or measured		
concentrations?		
Chemical method documented?	HPLC	
Concentration of carrier (if any) in	0.5 mL/L acetone	
test solutions		
Concentration 1 Nom; Meas (µg/L)	650; 620	2 reps, 10/rep
Concentration 2 Nom; Meas (µg/L)	1100; 650	2 reps, 10/rep
Concentration 3 Nom; Meas (µg/L)	1800; 1800	2 reps, 10/rep
Concentration 4 Nom; Meas (µg/L)	3000; 3200	2 reps, 10/rep
Concentration 5 Nom; Meas (µg/L)	5000; 4300	2 reps, 10/rep
Control	Negative: 0; 0	2 reps, 10/rep
	Solvent: 0; 0	2 reps, 10/rep
LC <sub>50</sub> (95% CI) (μg/L)	96 h: 1700 (1400-2000)	Method: probit or
		moving average

Notes: Prometryn solubility (S) = 31,250 ug/L  $\mu$ g/L, 2S = 62,500  $\mu$ g/L.

# Reliability points taken off for:

<u>Documentation:</u> Hardness (2), Alkalinity (2), Conductivity (2), Statistical significance (2), Significance level (2), Minimum significant difference (2), % control at NOEC/LOEC (2). Total: 100 - 14 = 86

<u>Acceptability:</u> Feeding (3), Hardness (2), Alkalinity (2), Conductivity (1), Random design (2), Adequate replication (2), Hypothesis tests (3). Total: 100 - 15 = 85

Reliability score: mean (86, 85) = 85.5

#### Carrassius auratus

Study: Beliles R P, Scott W Knott W. (1965) Prometryne: Safety evaluation on fish and wildlife (bobwhite quail, mallard duck, rainbow trout, sunfish, goldfish). Woodard Research Corporation. Presented to Geigy Agricultural Chemicals. CADPR study ID 92590.

RelevanceReliabilityScore: 90Score: 68Rating: RRating: L

Relevance points taken off for: Standard method (10). 100 - 10 = 90

	Beliles et al. 1965	C. auratus
Parameter	Value	Comment
Test method cited	Not reported	
Phylum/subphylum	Chordata	
Class	Actinopterygii	
Order	Cypriniformes	
Family	Cyprinidae	
Genus	Carrassius	
Species	auratus	
Family native to North America?	Yes	
Age/size at start of test/growth	Mean mass: 1.1 g	
phase	Mean length 3.5 cm	
Source of organisms	Hunting Creek Fisheries,	
	Thurmont, Maryland	
Have organisms been exposed to	No	
contaminants?		
Animals acclimated and disease-	Yes	10 d minimum
free?		
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	96 h	
Data for multiple times?	Yes	24, 48, 72, 96 h
Effect 1	Mortality	
Control response 1	Negative:	100 % survival
	24 h: 0	
	48 h: 0	
	72 h: 0	
	96 h: 0	
	Solvent:	
	24 h: 0	
	48 h: 0	
	72 h: 0	
	96 h: 0	
Temperature	17. 5 ± 1.5 °C	

	Beliles et al. 1965	C. auratus
Parameter	Value	Comment
Test type	Static renewal	24 h intervals
Photoperiod/light intensity	Not reported	
Dilution water	DI water	Reconstituted with CaSO <sub>4</sub> , MgSO <sub>4</sub> , NaHCO <sub>3</sub> , KCl
рН	Not reported	
Hardness	Not reported	
Alkalinity	Not reported	
Conductivity	Not reported	
Dissolved Oxygen	Not reported	Oxygenated prior to use
Feeding	Daily during acclimation only	Purina Trout Chow
Purity of test substance	99 %	
Concentrations measured?	Not reported	
Measured is what % of nominal?	Not reported	
Toxicity values calculated based on nominal or measured concentrations?	Not reported	
Chemical method documented?	Not reported	
Concentration of carrier (if any) in test solutions	Acetone, concentration not reported	Presumed 5 ppt as in solvent control
Concentration 1 Nom; Meas (µg/L)	560; not reported	2 reps, 5/rep
Concentration 2 Nom; Meas (µg/L)	1000; not reported	2 reps, 5/rep
Concentration 3 Nom; Meas (µg/L)	1800; not reported	2 reps, 5/rep
Concentration 4 Nom; Meas (µg/L)	3200; not reported	2 reps, 5/rep
Concentration 5 Nom; Meas (µg/L)	5600; not reported	2 reps, 5/rep
Concentration 6 Nom; Meas (µg/L)	10000; not reported	2 reps, 5/rep
Concentration 7 Nom; Meas (µg/L)	18000; not reported	2 reps, 5/rep
Control	Negative: 0; not reported Solvent: 0; not reported	2 reps, 5/rep Solvent: 5 ppt acetone
LC <sub>50</sub> (95% CI) (µg/L)	48 h: 9400 (5400-17000) 96 h: 3500 (530-6600)	Method: Litchfield and Wilcoxon

Notes: Prometryn solubility (S) = 31,250 ug/L  $\mu$ g/L, 2S = 62,500  $\mu$ g/L. All exposure concentrations were acceptable

#### Reliability points taken off for:

<u>Documentation:</u> Analytical method (4), Measured concentrations (3), Hardness (2), Alkalinity (2), Dissolved oxygen (4), Conductivity (2), pH (3), Photoperiod (3), Statistical significance (2), Significance level (2), Minimum significant difference (2), % control at NOEC/LOEC (2). Total: 100 - 31 = 69

<u>Acceptability:</u> Standard method (5), Measured concentrations within 20% nominal (4), Organisms randomized (1), Hardness (2), Alkalinity (2), Dissolved oxygen (6), Temperature

variation (3), Conductivity (1), Photoperiod (2), Random design (2), Minimum significant difference (1), Point estimates (3). Total: 100 - 32 = 67

Reliability score: mean (69, 67) = 68

### Cryptomonas sp.

Study: Leibig M., Schmidt G, Bontje D, Kooi BW, Streck G, Traunspurger W, Knacker T. (2008) Direct and indirect effects of pollutants on algae and algivorous ciliates in an aquatic indoor microcosm. *Aquatic Toxicology*. 88: 102-110.

RelevanceReliabilityScore: 85Score: 69.5Rating: LRating: L

Relevance points taken off for: Controls (15). 100 - 15 = 85

	Leibig et al. 2008	Cryptomonas sp.
Parameter	Value	Comment
Test method cited	Revised OECD guideline 201	
Phylum/subphylum, <i>Cryptomonas</i> sp.	Cryptophyta	
Order, Cryptomonas sp.	Cryptomonadales	
Family, <i>Cryptomonas</i> sp.	Cryptomonadaceae	
Genus, Cryptomonas sp.	Cryptomonas	
Family native to North America?	Yes	Ubiquitous
Age/size at start of test/growth phase	3 x 10 <sup>4</sup> cells mL <sup>-1</sup>	
Source of organisms	Strain SAG 26.80, Experimental Phycology and Culture Collection of Algae, Gottingen, Germany	Non-axenic
Have organisms been exposed to contaminants?	No	
Animals acclimated and disease-free?	Yes	
Animals randomized?	Yes	Aliquots of cell suspension
Test vessels randomized?	Not reported	-
Test duration	14 d	
Data for multiple times?	Yes	7, 14 d
Effect 1	Mean cell number	
Control response 1	Not reported	
Effect 2	Area below growth curve	
Control response 2	Not reported	
Effect 3	Growth rate	
Control response 3	Not reported	
Temperature	20 ± 1.5 °C	
Test type	Static	

	Leibig et al. 2008	Cryptomonas sp.
Parameter	Value	Comment
Photoperiod/light intensity	Continuous/33 ± 3 μmol	
	Continuous/33 ± 3 μmol photons m <sup>-2</sup> s <sup>-1</sup>	
Dilution water	Modified WC medium	Guillard and Lorenzen 1972
рН	$7.15 \pm 0.35$	
Hardness	Not reported	Growth medium
Alkalinity	Not reported	Growth medium
Conductivity	Not reported	Growth medium
Dissolved Oxygen	Not measured	
Feeding	Growth medium	
Purity of test substance	99.2	
Concentrations measured?	Yes	
Measured is what % of nominal?	± 20%	
Toxicity values calculated based on nominal or measured concentrations?	Measured	
Chemical method documented?	GC-MS	
Concentration of carrier (if any) in test solutions	Growth medium	
Concentration 1 Nom; Meas (µg/L)	Not reported; 6.9	4 reps, 3 x 10 <sup>4</sup> cells mL <sup>-1</sup> /rep
Concentration 2 Nom; Meas (µg/L)	Not reported; 10.3	Nominal concentration levels 1-5 in a geometric series from 7-35.4 µg/L
Concentration 3 Nom; Meas (µg/L)	Not reported; 15.5	Nominal concentration levels 1-5 in a geometric series from 7-35.4 µg/L
Concentration 4 Nom; Meas (µg/L)	Not reported; 23.2	Nominal concentration levels 1-5 in a geometric series from 7-35.4 µg/L
Concentration 5 Nom; Meas (µg/L)	Not reported; 34.8	Nominal concentration levels 1-5 in a geometric series from 7-35.4 µg/L
Control	0; 0	
EC <sub>50</sub> (95% CI) (μg/L)	Cell number: 7 d: 31.5 (29.5/34.1) Area under growth curve:	Method: ANOVA, probit

	Leibig et al. 2008	Cryptomonas sp.
Parameter	Value	Comment
	7 d: 22.9 (18.0/32.1)	
	Growth rate:	
	7 d: 39.3 (37.1/42.4)	
NOEC	Cell number:	Method: Student t-
	7 d: 23.2	test with Bonferroni
	14 d: 34.8	adjustment
	Area under growth curve:	p: 0.001
	7 d: 6.9	MSD: not reported
	14 d: 15.5	
	Growth rate:	
	7 d: 23.2	
	14 d: not calculable	
LOEC	Cell number:	
	7 d: 34.8	
	14 d: not calculable	
	Area under growth curve:	
	7 d: 10.3	
	14 d: 23.2	
	Growth rate:	
	7 d: 34.8	
	14 d: not calculable	
MATC (GeoMean NOEC, LOEC)	Cell number:	
	7 d: 24.8	
	14 d: not calculable	
	Area under growth curve:	
	7 d: 8.4	
	14 d: 19.0	
	Growth rate:	
	7 d: 28.4	
	14 d: not calculable	
% control at NOEC	Not calculable	
% control at LOEC	Not calculable	

Notes: Prometryn solubility (S) = 31,250 ug/L  $\mu$ g/L, 2S = 62,500  $\mu$ g/L so all exposure concentrations were acceptable.

#### Reliability points taken off for:

<u>Documentation</u>: Control type (8), Nominal concentrations (3), Dilution water (3), Hardness (2), Alkalinity (2), Dissolved oxygen (4), Conductivity (2), Minimum significant difference (2), % control at NOEC/LOEC (2). Total: 100 - 28 = 72

<u>Acceptability:</u> Appropriate control (6), Control response (9), Organisms randomized (1), Dilution water (2), Dissolved oxygen (6), Random design (2), Adequate replication (2), Dilution factor (2), Minimum significant difference (1), % control at NOEC (1), % control at LOEC (1). Total: 100 - 33 = 67

Reliability score: mean (72, 67) = 69.5

### Cyprinodon variegatus

Study: Surprenant DC. (1988b) Acute toxicity of prometryn technical to sheepshead minnow (*Cyprinodon variegatus*). Springborn Life Sciences, Inc., Wareham, MA. Laboratory study number 87-12-2588. Ciba-Geigy Corporation, Greensboro, NC. USEPA MRID 4057317.

RelevanceReliabilityScore: 85Score: 90.5Rating: LRating: R

Relevance points taken off for: Freshwater (15). 100 - 15 = 85

	Surprenant 1988b	C. variegatus
Parameter	Value	Comment
Test method cited	SLS protocol number	
	081787/CYP.SA (Appendix	
	I): "Protocol for conducting	
	a static acute toxicity test	
	with sheepshead minnow	
	(Cyprinodon variegatus)"	
Phylum/subphylum	Chordata	
Class	Actinopterygii	
Order	Cyprinodontiformes	
Family	Cyprinodontidae	
Genus	Cyprinodon	
Species	variegatus	
Family native to North America?	Yes	
Age/size at start of test/growth	Length: 29 mm	
phase	Weight: 0.42 g	
Source of organisms	Commercial supplier in	
	New Hampshire	
Have organisms been exposed to contaminants?	No	
Animals acclimated and disease-free?	Yes	14 d
Animals randomized?	Yes	
Test vessels randomized?	Not reported	
Test duration	96 h	
Data for multiple times?	Yes	24, 48, 72, 96 h
Effect 1	Mortality	
Control response 1	100 % survival	
Temperature	22 ± 1 °C	
Test type	Static	
Photoperiod/light intensity	25 footcandles	
Dilution water	Natural filtered seawater from Cape Cod Canal,	Salinity 32-35 ‰

	Surprenant 1988b	C. variegatus
Parameter	Value	Comment
	Bourne, Massachusetts	
pН	7.3-7.4	
Conductivity	21000 umhos/cm	
Dissolved Oxygen	88-92 %	
Feeding	Dry commercial pelleted	
	food <i>ad libitum</i> , daily until	
	test initiation	
Purity of test substance	98.1 %	
Concentrations measured?	Yes	
Measured is what % of nominal?	99-103	
Toxicity values calculated based on	Measured	
nominal or measured		
concentrations?		
Chemical method documented?	HPLC	
Concentration of carrier (if any) in	0.1 mL/L	Acetone
test solutions		
Concentration 1 Nom; Meas (µg/L)	1300; 880	1/rep, 10/rep
Concentration 2 Nom; Meas (µg/L)	2200; 1900	1/rep, 10/rep
Concentration 3 Nom; Meas (µg/L)	3600; 2800	1/rep, 10/rep
Concentration 4 Nom; Meas (µg/L)	6000; 4600	1/rep, 10/rep
Concentration 5 Nom; Meas (µg/L)	10000; 9100	1/rep, 10/rep
Control	Negative: 0;	1/rep, 10/rep
	Solvent: 0;	1/rep, 10/rep
LC <sub>50</sub> (95% CI) (µg/L)	24 h: > 9100	Method: probit
	48: > 9100	
	72 h: 8200 (5800-20100)	
	96 h: 5100 (4000-7000)	
NOEC	96 h: 880	Method: Not
		reported

Notes: Saltwater species.

Prometryn solubility (S) = 31,250 ug/L  $\mu$ g/L, 2S = 62,500  $\mu$ g/L.

# Reliability points taken off for:

<u>Documentation:</u> Statistical significance (2), Significance level (2), Minimum significant difference (2), % control at NOEC/LOEC (2). Total: 100 - 8 = 92

<u>Acceptability:</u> Hardness (2), Alkalinity (2), Conductivity (1), Random design (2), Adequate replication (2), % control at NOEC (1), % control at LOEC (1). Total: 100 - 11 = 89

Reliability score: mean (89, 92) = 90.5

# Chlorella vulgaris

Study: Ma J, Xu L, Wang S, Zheng R, Jin S, Huang S, and Huang Y. (2002) Toxicity of 40 herbicides to the green alga *Chlorella vulgaris*. *Ecotoxicology and environmental safety*, 51(2), 128-132.

RelevanceReliabilityScore: 75Score: 64Rating: LRating: L

Relevance points taken off for: Acceptable standard (10), Chemical purity (15). 100 - 25 = 75.

	Ma et al. 2002	C. vulgaris
Parameter	Value	Comment
Test method cited	Not reported	
Division	Chlorophyta	
Class	Trebouxiophyceae	
Order	Chlorellales	
Family	Chlorellaceae	
Genus	Chlorella	
Species	vulgaris	
Family native to North America?	Yes	
Age/size at start of test/growth	Algal cells, initial	
phase	concentration 8 x 10 <sup>5</sup> mL <sup>-1</sup>	
Source of organisms	Institute of Wuhan	
	Hydrobiology, Chinese	
	Academy of Science	
Have organisms been exposed to	No	
contaminants?		
Animals acclimated and disease-	Yes	
free?		
Animals randomized?	Yes	
Test vessels randomized?	Not reported	
Test duration	96 h	
Data for multiple times?	No	
Effect 1	Growth	
Control response 1	Not reported	
Temperature	$25 \pm 0$ °C	
Test type	Static	
Photoperiod/light intensity	Continuous/500 lx/cm <sup>-2</sup>	
Dilution water	HB-4 medium	Li, 1959
Feeding	Growth medium	
Purity of test substance	77.13 %	
Concentrations measured?	Not reported	
Measured is what % of nominal?	Not reported	
Toxicity values calculated based on	Nominal, although	

	Ma et al. 2002	C. vulgaris
Parameter	Value	Comment
nominal or measured concentrations?	concentrations not reported	
Chemical method documented?	No	
Concentration of carrier (if any) in test solutions	Distilled water, acetone, or methanol but not specified for prometryn	
Concentration Nom; Meas (µg/L)	Range of concentrations: 0-150, unspecified	3 reps, /rep
Control	Negative	
EC <sub>50</sub> (95% CI) (μg/L)	53.6	Method: Linear regression of transformed concentration as In data versus % inhibition p: 0.0031

Notes: Growth medium characteristics not reported. Chemical exposure concentrations not reported, only range given for linear regression analysis.

Prometryn solubility (S) = 31,250 ug/L,  $\mu\text{g/L}$ ,  $2\text{S} = 62,500 \mu\text{g/L}$ .

#### Reliability points taken off for:

<u>Documentation</u>: Chemical purity (5), Nominal concentrations (3), Measured concentrations (3), Hardness (2), Alkalinity (2), Dissolved oxygen (4), Conductivity (2), pH (3), Statistical significance (2), Significance level (2), Minimum significant difference (2), % control at NOEC/LOEC (2). Total: 100 - 32 = 68

Acceptability: Standard method (5), Control response (9), Chemical purity (10), Measured concentrations within 20% nominal (4), Carrier solvent (4), Dilution water (2), Hardness (2), Alkalinity (2), Dissolved oxygen (6), Temperature range (3), Conductivity (1), pH (2), Number of concentrations (3), Random design (2), Dilution factor (2), Minimum significant difference (1), % control at NOEC (1), % control at LOEC (1). Total: 100 - 60 = 40

Reliability score: mean (68, 60) = 64

### Daphnia magna

Study: Marchini S, Passerini L, Cesareo D, Tosato ML. (1988) Herbicidal triazines: acute toxicity on *Daphnia*, fish, and plants and analysis of its relationships with structural factors. *Ecotoxicology and Environmental Safety*. 16: 148-157.

RelevanceReliabilityScore: 92.5Score: 64.5Rating: RRating: L

Relevance points taken off for: Control response (7.5). 100 - 7.5 = 92.5

	Marchini et al. 1988	D. magna
Parameter	Value	Comment
Test method cited	OECD Guidelines No. 202	
Phylum/subphylum	Arthropoda/Crustacea	
Class	Branchiopoda	
Order	Cladocera	
Family	Daphniidae	
Genus	Daphnia	
Species	magna	
Family native to North America?	Yes	
Age/size at start of test/growth	< 24 h old	
phase		
Source of organisms	Laboratory of Comparative	Study's own lab
	Toxicology and	
	Ecotoxicology, Istituto	
	Superiore di Sanita, Rome,	
	Italy	
Have organisms been exposed to	No	
contaminants?		
Animals acclimated and disease-	Yes	
free?		
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	24, 48 h	
Data for multiple times?	Yes	24, 48 h
Effect 1	Immobilization	
Control response 1	Not reported	
Temperature	21 ± 1 °C	
Test type	Static	
Photoperiod/light intensity	12 l:12 d/Not reported	
Dilution water	Tap water, dechlorinated,	
	saturated with O <sub>2</sub>	
pН	8.4	
Hardness	250 mg/L CaCO <sub>3</sub>	

	Marchini et al. 1988	D. magna
Parameter	Value	Comment
Alkalinity	Within recommended limits	
Conductivity	Within recommended limits	
Dissolved Oxygen	Not reported	Dilution water
		saturated prior to
		use
Feeding	Chlorella	
Purity of test substance	> 96 %	
Concentrations measured?	Yes	
Measured is what % of nominal?	Not reported	
Toxicity values calculated based on	Not reported	
nominal or measured		
concentrations?		
Chemical method documented?	UV-Vis	
Concentration of carrier (if any) in	No carrier used	
test solutions		
Concentration 1 Nom; Meas (µg/L)	Number of test	4 reps, 5/rep
	concentrations and levels	
	not reported	
Control	Not reported	
EC <sub>50</sub> (95% CI) (μg/L)	24 h: 23,500	Method: Litchfiend
	48 h: 9,700	and Wilcoxon
		method (1949)

Notes:

Prometryn solubility (S) = 31,250 ug/L  $\mu$ g/L, 2S = 62,500  $\mu$ g/L.

Reliability points taken off for:

<u>Documentation</u>: Control type (8), Nominal concentrations (3), Measured concentrations (3), Dissolved oxygen (4), Hypothesis tests (8). Total: 100 - 31 = 74

<u>Acceptability:</u> Appropriate control (6), Control response (9), Measured concentrations within 20% nominal (4), Concentrations not > 2x solubility (4), Organisms randomized (1), Feeding (3), Exposure type (2), Dissolved oxygen (6), Number of concentrations (3), Random design (2), Dilution factor (2), Hypothesis tests (3). Total: 100 - 45 = 55

Reliability score: mean (74, 55) = 64.5

#### Lepomis macrochirus

Study: Beliles RP, Scott W, Knott W. (1965) Prometryne: Safety evaluation on fish and wildlife (bobwhite quail, mallard duck, rainbow trout, sunfish, goldfish). Woodard Research Corporation. Presented to Geigy Agricultural Chemicals. CADPR study ID 92590.

RelevanceReliabilityScore: 90Score: 68Rating: RRating: L

Relevance points taken off for: Standard method (10). 100 - 10 = 90

	Beliles et al. 1965	L. macrochirus
Parameter	Value	Comment
Test method cited	Not reported	
Phylum/subphylum	Chordata	
Class	Actinopterygii	
Order	Perciformes	
Family	Centrarchidae	
Genus	Lepomis	
Species	macrochirus	
Family native to North America?	Yes	
Age/size at start of test/growth	Mean mass: 0.8 g	
phase	Mean length 3.4 cm	
Source of organisms	Collected locally	
Have organisms been exposed to	Not reported	Collected locally
contaminants?		
Animals acclimated and disease-	Yes	10 d minimum
free?		
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	96 h	
Data for multiple times?	Yes	24, 48, 72, 96 h
Effect 1	Mortality	
Control response 1	Negative:	100 % survival
	24 h: 0	
	48 h: 0	
	72 h: 0	
	96 h: 0	
	Solvent:	
	24 h: 0	
	48 h: 0	
	72 h: 0	
	96 h: 0	
Temperature	$20.5 \pm 2.5  ^{\circ}\text{C}$	
Test type	Static renewal	24 h intervals

	Beliles et al. 1965	L. macrochirus
Parameter	Value	Comment
Photoperiod/light intensity	Not reported	
Dilution water	DI water	Reconstituted with
		CaSO <sub>4</sub> , MgSO <sub>4</sub> ,
		NaHCO <sub>3</sub> , KCl
рН	Not reported	
Hardness	Not reported	
Alkalinity	Not reported	
Conductivity	Not reported	
Dissolved Oxygen	Not reported	Oxygenated prior to use
Feeding	Daily during acclimation	Purina Trout Chow
Durity of test substance	only 99 %	
Purity of test substance Concentrations measured?	Not reported	
Measured is what % of nominal?	Not reported	
Toxicity values calculated based on	Not reported	
nominal or measured	Not reported	
concentrations?		
Chemical method documented?	Not reported	
Concentration of carrier (if any) in	Acetone, concentration not	Presumed 5 ppt as
test solutions	reported	in solvent control
Concentration 1 Nom; Meas (µg/L)	560; not reported	2 reps, 5/rep
Concentration 2 Nom; Meas (µg/L)	1000; not reported	2 reps, 5/rep
Concentration 3 Nom; Meas (µg/L)	1800; not reported	2 reps, 5/rep
Concentration 4 Nom; Meas (µg/L)	3200; not reported	2 reps, 5/rep
Concentration 5 Nom; Meas (µg/L)	5600; not reported	2 reps, 5/rep
Concentration 6 Nom; Meas (µg/L)	10000; not reported	2 reps, 5/rep
Concentration 7 Nom; Meas (µg/L)	18000; not reported	2 reps, 5/rep
Control	Negative: 0; not reported	2 reps, 5/rep
	Solvent: 0; not reported	Solvent: 5 ppt
		acetone
LC <sub>50</sub> (95% CI) (μg/L)	48 h: 14000 (9200, 21000)	Method: Litchfield
	96 h: 10000 (6200, 14000)	and Wilcoxon

Notes: Prometryn solubility (S) = 31,250 ug/L  $\mu$ g/L, 2S = 62,500  $\mu$ g/L. All exposure concentrations were acceptable

### Reliability points taken off for:

<u>Documentation:</u> Analytical method (4), Measured concentrations (3), Hardness (2), Alkalinity (2), Dissolved oxygen (4), Conductivity (2), pH (3), Photoperiod (3), Statistical significance (2), Significance level (2), Minimum significant difference (2), % control at NOEC/LOEC (2). Total: 100 - 31 = 69

<u>Acceptability:</u> Standard method (5), Measured concentrations within 20% nominal (4), Organisms randomized (1), Hardness (2), Alkalinity (2), Dissolved oxygen (6), Temperature variation (3), Conductivity (1), Photoperiod (2), Random design (2), Minimum significant difference (1), Point estimates (3). Total: 100 - 32 = 67

Reliability score: mean (69, 67) = 68

#### Mercenaria mercenaria

Study: Surprenant DC. (1988c) Acute Toxicity of prometryn technical to embryos-larvae of the quahog clam (*Mercenaria mercenaria*). Conducted by Springbron Life Sciences, Inc., Wareham, MA. Laboratory study number 88-1-2626. Ciba-Geigy Corporation, Greensboro, NC. USEPA MRID 40573719.

RelevanceReliabilityScore: 85Score: 82Rating: LRating: R

Relevance points taken off for: Freshwater (15). 100-15=85.

	Surprenant 1988c	M. mercenaria
Parameter	Value	Comment
Test method cited	EPA guideline reference	
	number 72-3; Proposed	
	Standard Practice for	
	Conducting Static Basic	
	Acute Toxicity Tests with	
	Larvae of Four Species of	
	Bivalve Molluscs (ASTM	
	1980, Draft No. 7)	
Phylum/subphylum	Mollusca	
Class	Bivalvia	
Order	Veneroida	
Family	Veneridae	
Genus	Mercenaria	
Species	mercenaria	
Family native to North America?	Yes	
Age/size at start of test/growth	Clam embryos/larvae	
phase		
Source of organisms	Aquacultural Research	
	Corporation, Dennis,	
	Massachusetts	
Have organisms been exposed to	No	
contaminants?		
Animals acclimated and disease-	Yes	
free?		
Animals randomized?	Yes	Not reported but
		assumed
		randomized because
		embryos so small
		that aliquots of
		specimens would be
		difficult to transfer

	Surprenant 1988c	M. mercenaria
Parameter	Value	Comment
		in a non- randomized manner
Test vessels randomized?	Not reported	
Test duration	48 h	
Data for multiple times?	No	
Effect 1	Normal larvae count	
Control response 1	Negative: 20,933	
	Solvent: 21,000	
Temperature	$22 \pm 1$ °C	
Test type	Static	
Photoperiod/light intensity	16 l: 8 d	
Dilution water	Filtered natural seawater,	5 μm
	Cape Cod Canal, Bourne,	Salinity: 31‰
	Massachusetts	
pН	7.9	
Dissolved Oxygen	6.4 mg/L	73 %
Feeding	Not reported	
Purity of test substance	98.1%	
Concentrations measured?	Yes	
Measured is what % of nominal?	51-39 %	
Toxicity values calculated based on	Measured	
nominal or measured		
concentrations?		
Chemical method documented?	HPLC	
Concentration of carrier (if any) in test solutions	0.5 mL/L	
Concentration 1 Nom; Meas (µg/L)	80,000; 31,000	25,920/rep, 3 reps
Concentration 2 Nom; Meas (µg/L)	48,000; 24,000	25,920/rep, 3 reps
Concentration 3 Nom; Meas (µg/L)	29,000; 16,000	25,920/rep, 3 reps
Concentration 4 Nom; Meas (µg/L)	17,000; 9,300	25,920/rep, 3 reps
Concentration 5 Nom; Meas (µg/L)	10,000; 5,100	25,920/rep, 3 reps
Controls	Negative: 0; < 1200	25,920//rep, 4 reps
	Solvent: 0; <1200	25,920//rep, 3 reps
EC <sub>50</sub> (95% CI) (μg/L)	48 h: 21,000 (120-51,000)	Method: Linear regression
NOEC	16,000	Method: ANOVA and Williams' Test p: 0.05 MSD: not reported
LOEC	24,000	The second secon
MATC	19595	
% control at NOEC	105 %	NOEC = 16,000
		22,133 (tmt) /
		20,971 (mean
		control) = 105 %

	Surprenant 1988c	M. mercenaria
Parameter	Value	Comment
% control at LOEC	13 %	LOEC = 24,000
		2667 (tmt) / 20,971
		(mean control) = 13
		%

Notes: Dilution water is seawater, used in accordance with USEPA/ASTM standard practices.

Prometryn solubility (S) = 31,250 ug/L  $\mu$ g/L, 2S = 62,500  $\mu$ g/L. All exposure concentrations were acceptable.

Reliability points taken off for:

<u>Documentation:</u> Hardness (2), Alkalinity (2), Conductivity (2), Statistics method (5), Hypothesis tests (8), Statistical significance (2), Minimum significant difference (2). Total: 100 - 23 = 77

<u>Acceptability:</u> Carrier solvent (4), Hardness (2), Alkalinity (2), Conductivity (1), Random design (2), Minimum significant difference (1), % control at LOEC (1). Total: 100 - 13 = 87

Reliability score: mean (77, 87) = 82

#### Oncorhynchus mykiss

Study: Beliles R P, Scott W, Knott W. (1965) Prometryne: Safety evaluation on fish and wildlife (bobwhite quail, mallard duck, rainbow trout, sunfish, goldfish). Woodard Research Corporation. Presented to Geigy Agricultural Chemicals. CADPR study ID 92590.

RelevanceReliabilityScore: 90Score: 68Rating: RRating: L

Relevance points taken off for: Standard method (10). 100 - 10 = 90

	Beliles et al. 1965	O. mykiss
Parameter	Value	Comment
Test method cited	Not reported	
Phylum/subphylum	Chordata	
Class	Actinopterygii	
Order	Salmoniformes	
Family	Salmonidae	
Genus	Oncorhynchus	
Species	mykiss	
Family native to North America?	Yes	
Age/size at start of test/growth	Mean mass: 0.9 g	
phase	Mean length 3.9 cm	
Source of organisms	Virginia Trout Company,	
	Inc., Monterey, Virginia	
Have organisms been exposed to	No	
contaminants?	***	10.1
Animals acclimated and disease-	Yes	10 d
free?	N. d. 1	
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	96 h	24 40 72 061
Data for multiple times?	Yes	24, 48, 72, 96 h
Effect 1	Mortality	100.0/
Control response 1	Negative:	100 % survival
	24 h: 0	
	48 h: 0	
	72 h: 0	
	96 h: 0	
	Solvent:	
	24 h: 0	
	48 h: 0	
	72 h: 0	
	96 h: 0	
Temperature	14.5 ± 1.5 °C	

	Beliles et al. 1965	O. mykiss
Parameter	Value	Comment
Test type	Static renewal	24 h intervals
Photoperiod/light intensity	Not reported	
Dilution water	DI water	Reconstituted with CaSO <sub>4</sub> , MgSO <sub>4</sub> , NaHCO <sub>3</sub> , KCl
рН	Not reported	
Hardness	Not reported	
Alkalinity	Not reported	
Conductivity	Not reported	
Dissolved Oxygen	Not reported	Oxygenated prior to use
Feeding	Daily during acclimation only	Purina Trout Chow
Purity of test substance	99 %	
Concentrations measured?	Not reported	
Measured is what % of nominal?	Not reported	
Toxicity values calculated based on nominal or measured	Not reported	
concentrations?		
Chemical method documented?	Not reported	
Concentration of carrier (if any) in	Acetone, concentration not	Presumed 5 ppt as
test solutions	reported	in solvent control
Concentration 1 Nom; Meas (µg/L)	560; not reported	2 reps, 5/rep
Concentration 2 Nom; Meas (µg/L)	870; not reported	2 reps, 5/rep
Concentration 3 Nom; Meas (µg/L)	1570; not reported	2 reps, 5/rep
Concentration 4 Nom; Meas (µg/L)	2780; not reported	2 reps, 5/rep
Concentration 5 Nom; Meas (µg/L)	4880; not reported	2 reps, 5/rep
Concentration 6 Nom; Meas (µg/L)	8730; not reported	2 reps, 5/rep
Control	Negative: 0; not reported	2 reps, 5/rep
	Solvent: 0; not reported	Solvent: 5 ppt
		acetone
LC <sub>50</sub> (95% CI) (μg/L)	48 h: 6200 (4100, 9400)	Method: Litchfield
	96 h: 2500 (1600, 4000)	and Wilcoxon

Notes: Prometryn solubility (S) = 31,250 ug/L  $\mu$ g/L, 2S = 62,500  $\mu$ g/L. All exposure concentrations were acceptable

#### Reliability points taken off for:

<u>Documentation</u>: Analytical method (4), Measured concentrations (3), Hardness (2), Alkalinity (2), Dissolved oxygen (4), Conductivity (2), pH (3), Photoperiod (3), Statistical significance (2), Significance level (2), Minimum significant difference (2), % control at NOEC/LOEC (2). Total: 100 - 31 = 69

<u>Acceptability:</u> Standard method (5), Measured concentrations within 20% nominal (4), Organisms randomized (1), Hardness (2), Alkalinity (2), Dissolved oxygen (6), Temperature variation (3), Conductivity (1), Photoperiod (2), Random design (2), Minimum significant difference (1), Point estimates (3). Total: 100 - 32 = 67

Reliability score: mean (69, 67) = 68

Procambarus fallax fallax virginalis

Study: Velisek, J. (2014) Effect of prometryne on early life stages of marbled crayfish (*Procambarus fallax f. virginalis*). Neuroendrocrinology Letters. 35: 93-98.

RelevanceReliabilityScore: 82.5Score: 80Rating: LRating: R

Relevance points taken off for: Standard method (10), Control response (7.5). 100 - 17.5 = 82.5

	Velisek 2014	P. fallax f. virginalis
Parameter	Value	Comment
Test method cited	Not reported	
Phylum/subphylum	Arthropoda/ Crustacea	
Class	Malacostraca	
Order	Decapoda	
Family	Cambaridae	
Genus	Procambarus	
Species	fallax fallax virginalis	
Family native to North America?	Yes	
Age/size at start of test/growth phase	Eggs, mean mass 2.27 mg, IX-X stage of embryonic development	From single marbled crayfish (female), carapace length 31.22 mm, postorbital carapace length 23.62, weight 9.19 g
Source of organisms	Cultured in laboratory	
Have organisms been exposed to contaminants?	No	
Animals acclimated and disease-free?	Yes	
Animals randomized?	Yes	
Test vessels randomized?	Not reported	
Test duration	53 d	
Data for multiple times?	No	
Effect 1	Mortality	
Control response 1	18 %	
Effect 2	Growth rate	
Control response 2	4.20	
Effect 3	Histopathology	Reported as percent inhibition of specific growth in data table
Control response 3	0%	
Effect 4	Body weight	

	Velisek 2014	P. fallax f. virginalis
Parameter	Value	Comment
Control response 4	24 d: 5.31 mg ± 0.42	
	53 d: 18.32 mg ± 5.34	
Temperature	$22.8 \pm 1.5  {}^{\circ}\text{C}$	
Test type	Static renewal	Renewed 3/wk until 24 d
Photoperiod/light intensity	11 1:13 d	
Dilution water	Tap water	
рН	7.5 - 8	
Hardness	Not reported	
Alkalinity	Not reported	
Conductivity	Not reported	
Dissolved Oxygen	> 60 %	
Feeding	1/d, ad libitum, brine shrimp	
Purity of test substance	99.3 %†	
Concentrations measured?	Yes	
Measured is what % of nominal?	≤ 10 %	
Toxicity values calculated based on	Not reported	
nominal or measured		
concentrations?	LCMCMG	
Chemical method documented?	LC-MS/MS	
Concentration of carrier (if any) in test solutions	Not reported	
	0.51	(0,, 1/, (, 1
Concentration 1 Nom; Meas (µg/L)	0.51; not reported	60 reps, 1/rep (single
		eggs in vessel to minimize
		contamination)
Company and a 2 Norman Manager (1)	144: not reported	
Concentration 2 Nom; Meas (µg/L)	144; not reported	60 reps, 1/rep (single eggs in vessel to
		minimize
		contamination)
Concentration 2 Name Mans (ug/L)	1444; not reported	60 reps, 1/rep (single
Concentration 3 Nom; Meas (µg/L)	1444, not reported	eggs in vessel to
		minimize
		contamination)
Concentration 4 Nom; Meas (µg/L)	4320; not reported	60 reps, 1/rep (single
Concentration 4 Norm, weas (µg/L)	1320, not reported	eggs in vessel to
		minimize
		contamination)
Control	Dilution water; not reported	60 reps, 1/rep (single
	, and the state of	eggs in vessel to
		minimize
		contamination)
LC <sub>50</sub> (95% CI) (μg/L)	Mortality, 53 d: 40	Method: probit
JU (>JU (MB/L)		r

Notes:

Prometryn solubility (S) = 31,250 ug/L  $\mu$ g/L, 2S = 62,500  $\mu$ g/L.

Reliability points taken off for:

<u>Documentation:</u> Measured concentrations (3), Hardness (2), Alkalinity (2), Conductivity (2), Hypothesis tests (8). Total: 100 - 17 = 83

<u>Acceptability:</u> Standard method (5), Hardness (2), Alkalinity (2), Temperature variation (3), Conductivity (1), Number of concentrations (3), Random design (2), Duration (2), Dilution factor (2), Hypothesis tests (3). Total: 100 - 23 = 75

Reliability score: mean (83, 75) = 79

Procambarus fallax fallax virginalis

Study: Velisek J. Effect of prometryne on early life stages of marbled crayfish (*Procambarus fallax f. virginalis*). 2014. *Neuroendrocrinology Letters*. 35: 93-98.

RelevanceReliabilityScore: 77.5Score: 80Rating: LRating: R

Relevance points taken off for: Endpoint linked to survival/growth/reproduction (15), Control response (7.5). 100-22.5=77.5

	Velisek 2014	P. fallax f. virginalis
Parameter	Value	Comment
Test method cited	Not reported	
Phylum/subphylum	Arthropoda/ Crustacea	
Class	Malacostraca	
Order	Decapoda	
Family	Cambaridae	
Genus	Procambarus	
Species	fallax	
Family native to North America?	Yes	
Age/size at start of test/growth phase	Eggs, mean mass 2.27 mg, IX-X stage of embryonic development	From single marbled crayfish (female), carapace length 31.22 mm, postorbital carapace length 23.62, weight 9.19 g
Source of organisms	Cultured in laboratory	8
Have organisms been exposed to contaminants?	No	
Animals acclimated and disease-free?	Yes	
Animals randomized?	Yes	
Test vessels randomized?	Not reported	
Test duration	53 d	
Data for multiple times?	No	
Effect 1	Mortality	
Control response 1	18 %	
Effect 2	Growth rate	
Control response 2	4.20	
Effect 3	Histopathology	Reported as percent inhibition of specific growth in data table
Control response 3	0%	

	Velisek 2014	P. fallax f. virginalis
Parameter	Value	Comment
Effect 4	Body weight	
Control response 4	$24 \text{ d: } 5.31 \text{ mg} \pm 0.42$	
	53 d: 18.32 mg ± 5.34	
Temperature	22.8 ± 1.5 °C	
Test type	Static renewal	Renewed 3/wk until 24 d
Photoperiod/light intensity	11 l:13 d	
Dilution water	Tap water	
pН	7.5 - 8	
Hardness	Not reported	
Alkalinity	Not reported	
Conductivity	Not reported	
Dissolved Oxygen	> 60 %	
Feeding	1/d, ad libitum, brine shrimp	
Purity of test substance	99.3 %†	
Concentrations measured?	Yes	
Measured is what % of nominal?	≤ 10 %	
Toxicity values calculated based on	Not reported	
nominal or measured		
concentrations?		
Chemical method documented?	LC-MS/MS	
Concentration of carrier (if any) in	Not reported	
test solutions	_	
Concentration 1 Nom; Meas (µg/L)	0.51; not reported	60 reps, 1/rep (single
, <u> </u>		eggs in vessel to
		minimize
		contamination)
Concentration 2 Nom; Meas (µg/L)	144; not reported	60 reps, 1/rep (single
		eggs in vessel to
		minimize
		contamination)
Concentration 3 Nom; Meas (µg/L)	1444; not reported	60 reps, 1/rep (single
		eggs in vessel to
		minimize
		contamination)
Concentration 4 Nom; Meas (µg/L)	4320; not reported	60 reps, 1/rep (single
		eggs in vessel to
		minimize
		contamination)
Control	Dilution water; not reported	60 reps, 1/rep (single
		eggs in vessel to
		minimize
		contamination)
LC <sub>50</sub> (95% CI) (μg/L)	Mortality, 53 d: 40	Method: probit
NOEC	0.1	Method: probit
		p: not reported

	Velisek 2014	P. fallax f. virginalis
Parameter	Value	Comment
		MSD: not reported
LOEC	0.51	
MATC (GeoMean NOEC, LOEC)	0.23	
% control at NOEC	Not calculable	
% control at LOEC	Not calculable	

Prometryn solubility (S) = 31,250 ug/L  $\mu$ g/L, 2S = 62,500  $\mu$ g/L.

Chronic toxicity values were not related to growth, reproduction, or mortality so points deducted. Acute toxicity value ( $LC_{50}$ ) related to mortality.

Reliability points taken off for:

<u>Documentation:</u> Measured concentrations (3), Hardness (2), Alkalinity (2), Conductivity (2), Hypothesis tests (8). Total: 100 - 17 = 83

<u>Acceptability:</u> Standard method (5), Hardness (2), Alkalinity (2), Temperature variation (3), Conductivity (1), Number of concentrations (3), Random design (2), Dilution factor (2), Hypothesis tests (3). Total: 100 - 23 = 77

Reliability score: mean (83, 77) = 80

#### Skeletonema costatum

Study: Hughes JS, Alexander MM. (1993) The toxicity of prometryn technical to *Skeleonema costatum*. Malcom Pirnie, Inc., White Plains, New York. Laboratory project ID B267-577-3. Ciba-Geigy Corporation, Greensboro, NC. USEPA MRID 42620202.

RelevanceReliabilityScore: 85Score: 98Rating: LRating: R

Relevance points taken off for: Freshwater (15). 100-15=85

	Hughes & Alexander 1993	S. costatum
Parameter	Value	Comment
Test method cited	MPI Protocol B267-577-3,	
	which satisfies EPA's	
	Pesticide Assessment	
	Guidelines	
Phylum/subphylum	Bacillariophyta	
Class	Coscinodiscophyceae/	
	Thalassiosirophycidae	
Order	Thalassiosirales	
Family	Skeletonemaceae	
Genus	Skeletonema	
Species	costatum	
Family native to North America?	Yes	
Age/size at start of test/growth	Algal cells	
phase		
Source of organisms	EPA Environmental	
	Research Laboratory, Gulf	
	Breeze, Florida	
Have organisms been exposed to	No	
contaminants?		
Animals acclimated and disease-	Yes	
free?		G: ·
Animals randomized?	Not reported	Given organism
		size and presence in
		growth medium, it is assumed that
		aliquots are
Test vessels randomized?	Yes	inherently randomly
Test duration	5 d	
Data for multiple times?	3, 4, 5 d	
Effect 1	Cell count	
Control response 1	3 d: 119280	
Control response 1	3 u. 117200	

	<b>Hughes &amp; Alexander 1993</b>	S. costatum
Parameter	Value	Comment
	4 d: 186665	
	5 d: 269270	
Temperature	20 ± 2 °C	
Test type	Static	
Photoperiod/light intensity	14:10, 1:d; 4306 lux	
Dilution water	Sterile synthetic seawater	Nutrients added
pH	8.1	
Feeding	Nutrients in seawater	
Purity of test substance	98.4 %	
Concentrations measured?	Yes	
Measured is what % of nominal?	83-103 %	
Toxicity values calculated based on	Measured	
nominal or measured		
concentrations?		
Chemical method documented?	GC	
Concentration of carrier (if any) in	Dimethylformamide, 0.5	
test solutions	mL/L	
Concentration 1 Nom; Meas (µg/L)	0.25; 0.259	10,000 cells/mL/rep
Concentration 2 Nom; Meas (µg/L)	0.625; 0.595	10/rep
Concentration 3 Nom; Meas (µg/L)	1.25; 1.16	10/rep
Concentration 4 Nom; Meas (µg/L)	2.5; 2.22	10/rep
Concentration 5 Nom; Meas (µg/L)	5; 4.54	10/rep
Concentration 6 Nom; Meas (µg/L)	10; 9.74	10/10/
Concentration 7 Nom; Meas (µg/L)	20; 18.8	
Control	Negative: 0; 0	10/rep
Control	Solvent: 0; 0	10/rep
EC <sub>25</sub> (95% CI) (μg/L)	4.98 (4.24-5.85)	Method:
EC <sub>25</sub> (95% CI) (µg/L)	7.63 (6.86-8.49)	Method: Nonlinear
Ευςο (7370 ετ) (μg/Ε)	7.03 (0.00 0.15)	regression
NOEC	2.22	Method: Dunnett's
11020		test
		p: 0.05
		MSD: Not reported
LOEC	4.54	
MATC	3.17	
% control at NOEC	3 d: 91 %	3 d: 108093 (tmt) /
	4 d: 95 %	119280 (mean
	5 d: 99 %	controls) = 91
		4 d: 178073 (tmt) /
		186665 (mean
		controls) = 95
		F 1 0/7570 /:
		5 d: 267573 (tmt) /
		269270 (mean

	Hughes & Alexander 1993	S. costatum
Parameter	Value	Comment
		controls) = 99
% control at LOEC	3 d: 60 %	3 d: 71120 (tmt) /
	4 d: 73 %	119280 (mean
	5 d: 89 %	controls) = 60
		4 d: 136763 (tmt) / 186665 (mean controls) = 73
		5 d: 238427 (tmt) / 269270 (mean
		controls) = 89

Notes:

Prometryn solubility (S) = 31,250 ug/L  $\mu$ g/L, 2S = 62,500  $\mu$ g/L.

Reliability points were not taken off for water quality parameters (hardness, alkalinity, conductivity) because there is no guidance for these parameters in the test guidelines for algal/plant studies, the growth medium used requires distilled water, and the medium is presumably appropriate for the test species because a specific culture media was used.

Reliability points taken off for:

<u>Documentation:</u> Minimum significant difference (2). Total: 100-2 = 98

Acceptability: Minimum significant difference (1), % control at LOEC (1). Total: 100-2 = 98

Reliability score: mean(98,98)=98

# Urotricha furcata

Study: Leibig M, Schmidt G, Bontje D, Kooi BW, Streck G, Traunspurger W, Knacker T. (2008) Direct and indirect effects of pollutants on algae and algivorous ciliates in an aquatic indoor microcosm. *Aquatic Toxicology*. 88: 102-110.

RelevanceReliabilityScore: 75Score: 64.5Rating: LRating: L

Relevance points taken off for: Standard method (10), Controls (15). 100 - 25 = 75

	Leibig et al. 2008	U. furcata
Parameter	Value	Comment
Test method cited	Not reported	
Phylum/subphylum, <i>U. furcata</i>	Ciliophora	
Class, U. furcata	Prostomatea	
Order, U. furcata	Prostomatida	
Family, <i>U. furcata</i>	Urotrichidae	
Genus, U. furcata	Urotricha	
Species, <i>U. furcata</i>	Furcata	
Family native to North America?	Yes	Ubiquitous
Age/size at start of test/growth	450 ciliates mL <sup>-1</sup>	-
phase		
Source of organisms	TH. Weisse, Austrian Academy of Sciences, Mondsee, Austria, derived from mesotrophic Lake Mondsee, Austria	Non-axenic.  In culture, flagellates serving as prey could not be removed without damaging cells so test started with minimum number of flagellates. Reduced light intensity prevented further growth of flagellates
Have organisms been exposed to contaminants?	U. furcata: originally derived from environment but aliquots from laboratory so assumed contaminant free	
Animals acclimated and disease-free?	Yes	

	Leibig et al. 2008	U. furcata
Parameter	Value	Comment
Animals randomized?	Yes	Aliquots of cell
		suspension
Test vessels randomized?	No	
Test duration	24 h	
Data for multiple times?	No	
Effect 1	Growth	
Control response 1	Not reported	Within 24 h control and concentration levels 1-3 growth occurred
Temperature	20 ± 1.5 °C	
Test type	Static	
Photoperiod/light intensity	Continuous/33 ± 3 μmol photons m <sup>-2</sup> s <sup>-1</sup>	
Dilution water	Modified WC medium	Guillard and Lorenzen 1972
pН	$7.15 \pm 0.35$	
Hardness	Not reported	Growth medium
Alkalinity	Not reported	Growth medium
Conductivity	Not reported	Growth medium
Dissolved Oxygen	> 90%	
Feeding	Growth medium	
Purity of test substance	99.2	
Concentrations measured?	Yes	
Measured is what % of nominal?	± 20%	
Toxicity values calculated based on nominal or measured concentrations?	Measured	
Chemical method documented?	GC-MS	
Concentration of carrier (if any) in test solutions	Growth medium	
Concentration 1 Nom; Meas (µg/L)	Five concentrations in geometric series from 940-1500	Unknown reps, 450 ciliates/rep
Control	Not reported	
NOEC	2200	Method: Student t- test with Bonferroni adjustment p: 0.001 MSD: not reported
LOEC	4500	•
MATC (GeoMean NOEC, LOEC)	3146	
% control at NOEC	Not calculable	
% control at LOEC	Not calculable	

Notes: Concentrations not reported, only geometric series range and number of concentrations tested. Little raw data reported for this species in this study.

Prometryn solubility (S) = 31,250 ug/L  $\mu$ g/L, 2S = 62,500  $\mu$ g/L.

Reliability points taken off for:

<u>Documentation</u>: Control type (8), Measured concentrations (3), Dilution water (3), Hardness (2), Alkalinity (2), Conductivity (2), Minimum significant difference (2), % control at NOEC/LOEC (2), Point estimates (8). Total: 100 - 32 = 68

<u>Acceptability:</u> Standard method (5), Appropriate control (6), Control response (9), Organisms randomized (1), Dilution water (2), Hardness (2), Alkalinity (2), Random design (2), Adequate replication (2), Dilution factor (2), Hypothesis tests (3), Point Estimates (3). Total: 100 - 39 = 61

Reliability score: mean (68, 61) = 64.5

# Appendix A5 - Aqueous studies rated N

#### Anabena variabilis

Study: Hawxby K, Tubea B, Ownby J, and Baslet E. (1977) Effects of various classes of herbicides on four species of algae. *Pesticide Biochemistry and Physiology*, 7, 203-299.

RelevanceReliabilityScore: 67.5Score: 53Rating: NRating: N

Relevance points taken off for: Acceptable method (10), Chemical purity (15), Controls described (7.5). 100 - 32.5 = 67.5

	Hawxby et al. 1977	A. variabilis
Parameter	Value	Comment
Test method cited	Not reported	
Phylum/subphylum	Cyanobacteria	
Order	Nostocales	
Family	Nostocaceae	
Genus	Anabena	
Species	variabilis	
Family native to North America?	Yes	
Age/size at start of test/growth	Cells used during	
phase	exponential growth phase	
Source of organisms	Carolina Biological Supply	Unialgal but not axenic
Have organisms been exposed to contaminants?	No	
Animals acclimated and disease-free?	Yes	
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	48 h	
Data for multiple times?	No	
Effect 1	Growth	Via optical density
Control response 1	Not reported	
Effect 2	Growth rate	
Control response 2	Not reported	
Effect 3	Photosynthetic rate	
Control response 3	Not reported	
Effect 4	Endogenous respiration	
Control response 4	Not reported	
Temperature	25 ± 1.0 °C	
Test type	Static	
Photoperiod/light intensity	Continuous/3200 lux	
Dilution water	Not reported	
рН	Not reported	Growth medium pH

	Hawxby et al. 1977	A. variabilis
Parameter	Value	Comment
		= 6.6
Hardness	Not reported	
Alkalinity	Not reported	
Conductivity	Not reported	
Dissolved Oxygen	Not reported, vessels	
	bubbled with air	
	continuously	
Feeding	Growth medium	Bold's basal
		medium
Purity of test substance	Not reported	
Concentrations measured?	No	
Measured is what % of nominal?	Not applicable	
Toxicity values calculated based on	Nominal	
nominal or measured		
concentrations?		
Chemical method documented?	Not applicable	
Concentration of carrier (if any) in	Not reported	
test solutions		
Concentration 1 Nom; Meas (µg/L)	24.13 (0.1 μM)	9 reps, 9 mL/rep
Concentration 2 Nom; Meas (µg/L)	241.3 (1.0 μM)	9 reps, 9 mL/rep
Concentration 3 Nom; Meas (µg/L)	2413 (10.0 μM)	9 reps, 9 mL/rep
Control	0	9 reps, 9 mL/rep
EC <sub>50</sub> (95% CI) (μg/L)	Growth: 3	Method:
	Photosynthesis: 0.17	Graphically after
		Duncan's multiple
		range test

Notes: Non-standard species used.

Solubility (S) =  $31,250 \mu g/L$ ,  $2S = 62,500 \mu g/L$ .

#### Reliability points taken off for:

<u>Documentation</u>: Control type (8), Chemical purity (5), Analytical method (4), Measured concentrations (3), Dilution water (3), Hardness (2), Alkalinity (2), Dissolved oxygen (4), Conductivity (2), Minimum significant difference (2), Point estimates (8). Total: 100 – 43 = 57

Acceptability: Standard method (5), Appropriate control (6), Chemical purity (10), Measured concentrations within 20% nominal (4), Organisms randomized (1), Adequate organisms per rep (2), Dilution water (2), Hardness (2), Alkalinity (2), Dissolved oxygen (6), Conductivity (1), Number of concentrations (3), Random design (2), Dilution factor (2), Minimum significant difference (1), % control at NOEC (1), % control at LOEC (1). Total: 100 - 51 = 49

Reliability score: mean (57, 49) = 53

## Chlorella pyrenoidosa

Study: Hawxby K, Tubea B, Ownby J, and Baslet E. (1977) Effects of various classes of herbicides on four species of algae. *Pesticide Biochemistry and Physiology*, 7, 203-299.

RelevanceReliabilityScore: 67.5Score: 53Rating: NRating: N

Relevance points taken off for: Acceptable method (10), Chemical purity (15), Controls described (7.5). 100 - 32.5 = 67.5

	Hawxby et al. 1977	C. pyrenoidosa
Parameter	Value	Comment
Test method cited	Not reported	
Division	Chlorophyta	
Class	Trebouxiophyceae	
Order	Chlorellales	
Family	Chlorellaceae	
Genus	Chlorella	
Species	pyrenoidosa	
Family native to North America?	Yes	
Age/size at start of test/growth	Cells used during	
phase	exponential growth phase	
Source of organisms	Carolina Biological Supply	Unialgal but not axenic
Have organisms been exposed to contaminants?	No	
Animals acclimated and disease-free?	Yes	
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	24 h	
Data for multiple times?	No	
Effect 1	Growth	Via optical density
Control response 1	Not reported	
Effect 2	Growth rate	
Control response 2	Not reported	
Effect 3	Photosynthetic rate	
Control response 3	Not reported	
Effect 4	Endogenous respiration	
Control response 4	Not reported	
Temperature	25 ± 1.0 °C	
Test type	Static	
Photoperiod/light intensity	Continuous/3200 lux	
Dilution water	Not reported	

	Hawxby et al. 1977	C. pyrenoidosa
Parameter	Value	Comment
pH	Not reported	Growth medium pH
		= 6.6
Hardness	Not reported	
Alkalinity	Not reported	
Conductivity	Not reported	
Dissolved Oxygen	Not reported, vessels	
	bubbled with air	
	continuously	
Feeding	Growth medium	Bold's basal
		medium
Purity of test substance	Not reported	
Concentrations measured?	No	
Measured is what % of nominal?	Not applicable	
Toxicity values calculated based on	Nominal	
nominal or measured		
concentrations?		
Chemical method documented?	Not applicable	
Concentration of carrier (if any) in	Not reported	
test solutions		
Concentration 1 Nom; Meas (µg/L)	24.13 (0.1 μM)	9 reps, 9 mL/rep
Concentration 2 Nom; Meas (µg/L)	241.3 (1.0 μM)	9 reps, 9 mL/rep
Concentration 3 Nom; Meas (µg/L)	2413 (10.0 μM)	9 reps, 9 mL/rep
Control	0	9 reps, 9 mL/rep
EC <sub>50</sub> (95% CI) (μg/L)	Growth: 241.3 (1.0μM)	Method:
	Photosynthesis: 241.3	Graphically after
	$(1.0\mu\text{M})$	Duncan's multiple
		range test

Notes: Non-standard species used.

Solubility (S) =  $31,250 \mu g/L$ ,  $2S = 62,500 \mu g/L$ .

#### Reliability points taken off for:

<u>Documentation</u>: Control type (8), Chemical purity (5), Analytical method (4), Measured concentrations (3), Dilution water (3), Hardness (2), Alkalinity (2), Dissolved oxygen (4), Conductivity (2), Minimum significant difference (2), Point estimates (8). Total: 100 - 43 = 57

<u>Acceptability:</u> Standard method (5), Appropriate control (6), Chemical purity (10), Measured concentrations within 20% nominal (4), Organisms randomized (1), Adequate organisms per rep (2), Dilution water (2), Hardness (2), Alkalinity (2), Dissolved oxygen (6), Conductivity (1), Number of concentrations (3), Random design (2), Dilution factor (2), Minimum significant difference (1), % control at NOEC (1), % control at LOEC (1). Total: 100 - 51 = 49

Reliability score: mean (57, 49) = 53

## Chlorella pyrenoidosa

Study: J Ma, W Liang, L Xu, S Wang, Y Wei, J Lu. (2001) Acute Toxicity of 33 Herbicides to the Green Alga Chlorella pyrenoidosa. *Bulletin of Environmental Contamination and Toxicology*, 66:536–541.

RelevanceReliabilityScore: 60Score: 38.5Rating: NRating: N

Relevance points taken off for: Acceptable standard (or equivalent) method used (10), Chemcial purity (15), Controls-Described (i.e., solvent, dilution water, etc.) (7.5), Controls-Response reported and meets acceptability requirements (7.5). Total: 100-25 = 75.

	Ma et al. 2001	C. pyrenoidosa
Parameter	Value	Comment
Test method cited	None	
Phylum/subphylum	Chlorophyta	
Class	Trebouxiophyceae	
Order	Chlorellales	
Family	Chlorellaceae	
Genus	Chlorella	
Species	pyrenoidosa	
Family native to North America?	Yes	
Age/size at start of test/growth phase	Algal cells, 6 x 10 <sup>5</sup> cells/mL	
Source of organisms	Laboratory culture	Institute of Wuhan Hydrobiology, Chinese Academy of Science
Have organisms been exposed to contaminants?	No	
Animals acclimated and disease-free?	Yes	
Animals randomized?	Not reported	
Test vessels randomized?	Not reported	
Test duration	96 hours	
Data for multiple times?	No	
Effect 1	Growth	
Control response 1	Not reported	
Temperature	25 °C	
Test type	Static	
Photoperiod/light intensity	Continuous @ 5000 lux/cm <sup>2</sup>	
Dilution water	Liquid HB-4 medium	
рН	Not reported	
Hardness	Not reported	

	Ma et al. 2001	C. pyrenoidosa
Parameter	Value	Comment
Alkalinity	Not reported	
Conductivity	Not reported	
Dissolved Oxygen	Not reported	
Feeding	Growth medium not renewed	
Purity of test substance	77%	
Concentrations measured?	No	
Measured is what % of nominal?	Not applicable	
Toxicity values calculated based on nominal or measured concentrations?	Nominal	
Chemical method documented?	Not applicable	
Concentration of carrier (if any) in test solutions	Not reported	
Concentration 1 Nom; Meas (µg/L)	Not reported	reps, /rep
Concentration 2 Nom; Meas (µg/L)	Not reported	
Concentration 3 Nom; Meas (µg/L)	Not reported	
Concentration 4 Nom; Meas (µg/L)	Not reported	
Concentration 5 Nom; Meas (µg/L)	Not reported	
Concentration 6 Nom; Meas (µg/L)	Not reported	
Control	Not described	
EC <sub>50</sub> (95% CI) (μg/L)	12	Method: calculated via spectrophotometric count

Notes: No control data reported. Multiple herbicides tested with various solvents used, although unspecified as to which was used for which herbicide. Control solvent not reported.

EPA guidance recommends algal species for testing, with *C. pyrenoidosa* not being one of them (alternate).

#### Reliability points taken off for:

<u>Documentation:</u> Control type (8), Organism life stage/size (5), Analytical method (4), Measured concentrations (3), Dilution water (3), Hardness (2), Alkalinity (2), Dissolved oxygen (4), Conductivity (2), pH (3), Methods identified (5), Statistical significance (2), Minimum significant difference (2), % control at NOEC/LOEC (2). Total: 100 - 47 = 53

Acceptability: Standard method (5), Appropriate control (6), Control response (9), Measured concentrations within 20% nominal (4), Concentrations not > 2x solubility (4), Carrier solvent (4), Appropriate size/age/growth phase (3), Organisms randomized (1), Adequate organisms per rep (2), Acclimation (1), Dilution water (2), Hardness (2), Alkalinity (2), Dissolved oxygen (6), Temperature variation (3), Conductivity (1), pH (2), Photoperiod (2), Number of concentrations (3), Random design (2), Adequate replication (2), Dilution factor (2), Statistical method (2), Hypothesis tests (3), Minimum significant difference (1), % control at NOEC (1), % control at LOEC (1). Total: 100 - 76 = 24

Reliability score: mean(53,24) = 38.5

#### Raphidocelis subcapitata

Study: Ma J, Wang S, Ma L, Chen X, Xu R. (2006) Toxicity assessment of 40 herbicides to the green alga *Raphidocelis subcapitata*. *Ecotoxicology and Environmental Safety*, 63, 456-462.

RelevanceReliabilityScore: 67.5Score: 72Rating: NRating: L

Relevance points taken off for: Standard method (10), Chemical purity (15), Control response (7.5). 100 - 32.5 = 67.5

	Ma et al. 2006	R. subcapitata
Parameter	Value	Comment
Test method cited	Not reported	
Phylum/subphylum	Chlorophyta	
Class	Chlorophyceae	
Order	Sphaeropleales	
Family	Selenastraceae	
Genus	Raphidocelis	
Species	subcapitata	
Family native to North America?	Yes	
Age/size at start of test/growth	Exponential	
phase		
Source of organisms	Institute of Wuhan	
	Hydrobiology, Chinese	
	Academy of Science	
Have organisms been exposed to	No	
contaminants?		
Animals acclimated and disease-	Yes	
free?		
Animals randomized?	Yes	15 mL aliquots
Test vessels randomized?	Not reported	
Test duration	96 h	
Data for multiple times?	No	
Effect 1	Growth	
Control response 1	Not reported	
Temperature	25 °C	Range not reported
Test type	Static	
Photoperiod/light intensity	450 E m <sup>-2</sup> s <sup>-1</sup>	
Dilution water	Growth medium	Prepared with
		distilled water;
		Chinese National
		Environmental
		Protection Agency
		Guidelines 201,

	Ma et al. 2006	R. subcapitata
Parameter	Value	Comment
		HB-4 medium
pH	Not reported	
Hardness	Not reported	
Alkalinity	Not reported	
Conductivity	Not reported	
Dissolved Oxygen	Not reported	
Feeding	Growth medium	
Purity of test substance	77 %	Technical product
Concentrations measured?	Not reported	
Measured is what % of nominal?	Not reported	
Toxicity values calculated based on	Not reported	
nominal or measured		
concentrations?		
Chemical method documented?	Not reported	
Concentration of carrier (if any) in	Acetone (< 0.05% in	
test solutions	medium) or distilled water	
Concentrations Nom; Meas (µg/L)	Not reported; "A wide range	$3 \text{ reps}, 5 \times 10^4$
	of concentrations" was	cells/rep
	tested	
Control	0, not reported	
EC <sub>50</sub> (95% CI) (μg/L)	11.7	Method: Linear
		regression analysis
		of transformed
		herbicide
		concentration as
		natural log data vs.
		% inhibition

Notes:

Prometryn solubility (S) = 31,250 ug/L  $\mu$ g/L, 2S = 62,500  $\mu$ g/L.

Reliability points were not taken off for water quality parameters (hardness, alkalinity, conductivity) because there is no guidance for these parameters in the test guidelines for algal/plant studies, the growth medium used requires distilled water, and the medium is presumably appropriate for the test species because a specific culture media was used.

#### Reliability points taken off for:

<u>Documentation</u>: Analytical method (4), Nominal concentrations (3), Measured concentrations (3), Minimum significant difference (2), % control at NOEC/LOEC (2). Total: 100 - 14 = 86

<u>Acceptability:</u> Standard method (5), Control response (9), Chemical purity (10), Measured concentrations within 20% nominal (4), Concentrations not > 2x solubility (4), Temperature range (3), Random design (2), Dilution factor (2), Minimum significant difference (1), % control at NOEC (1), % control at LOEC (1). Total: 100 - 42 = 58

Reliability score: mean (86, 58) = 72